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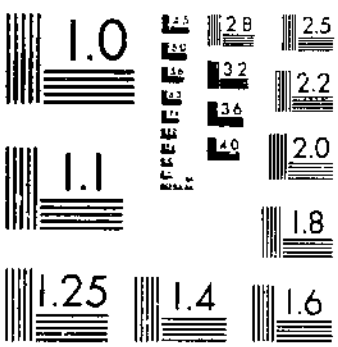

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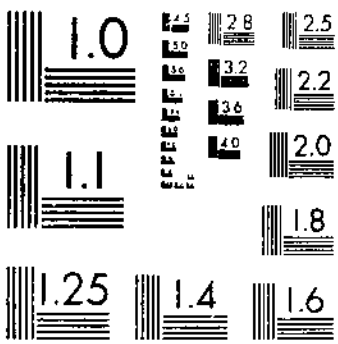

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NATIONAL BUREAU OF STANDARDS-1963-A

TECHNICAL BULLETIN No. 64 MARCH, 1928

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BACTERIOLOGY AND CHEMISTRY OF OYSTERS

WITH SPECIAL REFERENCE TO REGULATORY CONTROL OF PRODUCTION, HANDLING, AND SHIPMENT

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INTRODUCTION

The production, handling, and shipping of oysters were always of interest to the Bureau of Chemistry in its enforcement of the Federal food and drugs act. The possibility of pollution with sewage and trade waste, the rapidity with which spoilage takes place when

[1] The writers are particularly indebted to Payn B. Parsons, formerly of the Bureau of Chemistry, now with the New York State Conservation Commission, for many suggestions and much material, especially on the bacteriology of oysters. H. D. Pease, of the Pease Laboratories, F. P. Gorham, of Brown University, and L. A. Round and S. DeM. Gage, both of the Rhode Island State Board of Health Laboratories, have very kindly helped with suggestions and advice. R. W. Balcom, of the Food, Drug, and Insecticide Administration, assisted in planning and preparing the bulletin. Finally, acknowledgment is made to a large number of individuals in the service whose identity could not be learned from the official reports of their analyses which have been used in this bulletin.

Although this bulletin is published after the establishment of the Bureau of Chemistry and Soils and the Food, Drug, and Insecticide Administration, it is a contribution from the old Bureau of Chemistry rather than from either of the two new units.

oysters are handled improperly, and the likelihood of adulteration with water during the washing processes have made necessary extensive biological and chemical investigations to obtain the information required to establish regulations controlling the shipment of oysters in interstate commerce. The Federal food and drugs act (*91*)[2] is applicable to oysters in that it defines adulteration as (*a*) the mixing of any substance with a food product so as to reduce or lower or injuriously affect its quality or strength, (*b*) the substitution of any substance wholly or in part for the article, (*c*) the removal of any valuable constituent wholly or in part from the food product, (*d*) the presence of any poisonous or other added deleterious ingredient which renders the food injurious to health, and (*e*) the presence of any filthy, decomposed, or putrid animal or vegetable substance. Definitions of misbranded products given in section 8 of the act are particularly applicable to canned oysters.

To obtain reliable data upon the many biological and chemical problems surrounding the production and shipment of oysters, upon which to base interpretations of the food and drugs act and rulings that are just to both the producer and the consumer, the Bureau of Chemistry conducted many investigations to determine the biological principles of self-purification of oysters, the purification by treatment with calcium hypochlorite, the hibernation of oysters, and the effect of various methods of washing and shipping.

This bulletin presents a survey of the results of such investigations, supplemented where necessary by references to the findings of other investigators.

THE OYSTER INDUSTRY

Mollusks of the group that includes oysters are called bivalves, because they have two shells or valves. Oysters of commerce belong to the genus Ostrea, a great many species of which are grown in various parts of the world. Thus the oyster produced in Europe is the *Ostrea edulis* and the principal species in Japan is the *O. cucullata*. Two species are found on the coasts of Mexico, *O. elongata* on the eastern coast and *O. columbiensis* on the western coast. *O. puelchana* is produced on the eastern coast of South America and *O. columbiensis* and *O. chilensis* on the western coast. Other species range from Norway to Chile and New Zealand.

In the United States the principal oyster of commerce is the Atlantic and Gulf coast oyster, *O. virginica*. The small Olympia oyster, *O. lurida*, is grown in the waters of the Pacific Northwest, especially in and about Puget Sound and Willapa Harbor off the coast of Washington.

Oysters are produced commercially in all the Atlantic and Gulf Coast States except Maine and New Hampshire. On the Pacific coast, Oregon, Washington, and California produce for the market either the native Olympia oyster or the eastern oyster, and sometimes both (*55*).

Churchill, in discussing the geographical distribution of oyster-growing areas, has described in some detail the sections where oysters

[2] Reference is made by italic numbers in parenthesis to "Literature cited," p. 70.

are grown (*26*). Most of the oysters on the Atlantic and Gulf coasts are grown in rivers and bays, and in coves along the shore.

The methods of cultivating, harvesting, and handling oysters vary somewhat in the different regions. In certain sections oyster beds are leased from the State and the oysters are cultivated by the lessee. In other sections oysters grow naturally on beds or reefs and can be taken by anyone who has the proper license. Different types of gear are used; different classes of employees are hired to do the work; different shucking and washing methods exist. Furthermore, the conditions of pollution are different in various localities and, owing to the difference in the salinity of the waters, there are differences in the chemical composition of the oysters. No one investigation to fix regulations controlling the oyster industry can apply to all localities. It has been necessary to make extensive studies of conditions in all oyster-producing regions in order to reach just decisions upon regulatory matters.

The methods of gathering, storing, shucking, washing, and marketing oysters are described in detail by Churchill (*26*). Although oysters are usually shipped raw, some are canned. Canned oyster juice and dried oysters are sold in limited quantities.

The total production of oysters for the market in the United States reaches several million bushels annually. Statistics are not available for the oyster crop for the United States for any one year. Table 1, which gives the number of bushels of oysters grown for market by each producing State on the Atlantic, Gulf, and Pacific coasts, shows that Maryland and Virginia, drawing upon Chesapeake Bay for their supply, produce many more oysters than any other region. The Pacific coast production appears to be rather small as compared with that of some of the Atlantic and Gulf coast regions.

TABLE 1.—*Oyster production of the United States* [1]

State	Quantity [2]	Value	State	Quantity [1]	Value
Atlantic coast:	*Bushels*	*Dollars*	Atlantic and Gulf coasts:	*Bushels*	*Dollars*
Massachusetts [3]	110,002	279,547	Florida [6]	308,344	86,309
Rhode Island [3]	894,537	1,242,535	Gulf coast:		
Connecticut [4]	709,046	403,660	Alabama [6]	233,080	86,719
New York [4]	1,346,210	1,775,453	Mississippi [6]	1,606,422	472,652
New Jersey [4]	1,578,165	2,070,496	Louisiana [6]	1,022,109	770,434
Delaware [4]	401,826	368,115	Texas [6]	359,978	176,070
Maryland [5]	4,547,471	2,291,120	Pacific coast:		
Virginia [5]	3,225,844	2,167,923	Washington [7]	60,484	368,463
North Carolina [6]	559,898	220,576	Oregon [7]	19,714	7,500
South Carolina [6]	748,906	113,855	California [7]	13,514	101,351
Georgia [6]	245,762	86,771			

[1] Statistics furnished by Bureau of Fisheries, U. S. Department of Commerce.
[2] This represents market oysters. It is exclusive of seed oysters produced.
[3] For 1919.
[4] For 1921.
[5] For 1920.
[6] For 1923.
[7] For 1922.

Table 2 shows the number of cases of canned oysters produced by various States in 1924, with the total value of the output.

TABLE 2.—*Number of cases and value of canned oysters*[1] *produced in the United States in 1924* (97)

Cans in each case		Number of cases produced in—								Total	
Size	Number	Maryland	North Carolina	South Carolina	Georgia	Florida	Alabama	Mississippi	Louisiana and Texas	Cases	Value
Ounces											
4	48	7,343	668	[2] 12,855				32,025		52,901	$202,432
5	48	40,715	32,221	72,387	12,120	12,608	9,922	120,180	7,585	307,747	1,052,810
6	48	7,084		104					2,000	9,788	97,080
8	24	3,138		2,105		278		16,730		22,341	110,558
10	24	9,692	3,969	16,267	100	400	1,500	35,594		66,027	347,236
12	24	723								723	7,025
Total		68,695	36,857	103,808	12,220	13,286	11,422	204,548	9,585	460,427	2,478,044

[1] Oysters were canned at 9 plants in Maryland, 6 in North Carolina, 13 in South Carolina, 6 in Georgia, 6 in Florida, 5 in Alabama, 21 in Mississippi, 6 in Louisiana, and 1 in Texas.
[2] Includes pack of 3-ounce cans converted to the equivalent of 4-ounce cans, 4 dozen to the case.

In order to indicate the relative value of the oyster crop as compared with that of the entire fisheries output of each oyster producing region, Table 3 has been compiled. In compiling the data for this table oysters from private beds and from public beds were added together. No oysters produced for seed were considered.

TABLE 3.—*Oyster output and total fisheries output by sections*[1]

States	Year	Total fisheries	Oysters[2]	Part represented by oysters	Total value of fisheries	Value of oysters	Value represented by oysters
		Pounds	*Pounds*	*Per cent*	*Dollars*	*Dollars*	*Per cent*
Massachusetts, Rhode Island, and Connecticut	1919	318,854,771	11,999,715	3.8	15,856,962	2,015,798	12.7
New York, New Jersey, and Delaware	1921	332,337,129	23,253,407	7.0	11,622,772	4,214,964	36.2
Maryland and Virginia	1920	630,740,884	54,413,205	10.3	12,740,392	4,459,643	35.0
North Carolina, South Carolina, Georgia, and Florida (east coast)	1923	228,747,930	11,172,836	4.9	5,087,340	448,137	8.8
Florida (west coast), Alabama, Mississippi, Louisiana, and Texas	1923	160,324,012	24,823,309	15.5	8,096,050	1,574,445	19.4
Washington, Oregon, and California	1922	282,968,421	[3] 592,084	.2	12,983,583	417,314	3.2

[1] Compiled from U. S. Dept. Commerce, Bur. Fisheries Stat. Buls. (*92, 93, 94, 95, 96, 98*).
[2] Market oysters, exclusive of seed oysters.
[3] Includes both eastern and native oysters.

CHEMICAL COMPOSITION OF OYSTERS

MEATS AND LIQUOR

Very few results showing the exact chemical composition of oysters are available. For enforcing the Federal food and drugs act and for studying the changes that take place during floating (p. 36) and washing (p. 43), it has been sufficient to determine the solids, ash, and salt content. A large number of analyses indicating the proportion of solids and salt in normal oysters from various localities are therefore at hand.

From the few detailed analyses available it is evident that oysters are very complex bodies, high in nitrogen and phosphorus-contain-

ing compounds. No doubt, both water-soluble and water-insoluble proteins are present. Carbohydrates, in the form of glycogen, are found in varying quantities. Oysters also contain fatty bodies, which are usually reported as "ether extract." The ash of oysters contains, besides sodium chloride, probably almost every chemical element of sea water. Iodine and traces of bromine are present, as well as calcium and magnesium carbonates.

According to an anonymous review (*1*), the average weight of the native English oyster is 142 grains, the moisture content is between 77 and 83 per cent, the oyster contains organic matter up to 21 per cent, and the mineral matter content is between 1.6 and 2.5 per cent.

Protein constitutes 46.3 per cent of the organic matter examined, glycogen being present to the extent of 4 per cent, and the fat content is 4.7 per cent. The rest of the organic portion is composed of nonnitrogenous matter. Glycero-phosphoric compounds, such as lecithin and glycero-phosphates of alkali metals, are present. Fifty per cent of the mineral matter is composed of soluble phosphates and 32 per cent is sodium chloride. There are smaller proportions of magnesium and calcium phosphates, with traces of copper, zinc, and other metals.

Mitchell (*62*) reported that the carbohydrate glycogen is the substance the presence or absence of which makes oysters fat or lean. He found that the proportions of glycogen in 32 determinations varied from 3.05 to 22.46 per cent of the ash-free solids. When food is scarce oysters use glycogen to spare the proteins.

In analyses of dried samples Hindman and Goodrich (*45*) found that 7.4 per cent protein, 1.62 per cent fat, 2.12 per cent ash, and 3.2 per cent carbohydrate were present in Atlantic coast oysters; and 7.58 per cent protein, 1.64 per cent fat, 2.07 per cent ash, and 3.4 per cent carbohydrate in Puget Sound oysters. These data are in good agreement with the results of analyses made in the Office of Experiment Stations, United States Department of Agriculture (*13*), which showed that oysters contain 13.1 per cent solids, 6.2 per cent protein, 1.2 per cent fat, 3.7 per cent carbohydrates, and 2 per cent ash. Maximum, minimum, and average figures from the very large number of analyses made by the Bureau of Chemistry (Table 4) show the range of solids, ash, and salt in oysters from known sources.

TABLE 4.—*Solids, ash, and salt content of authentic samples of oysters*

Source and condition of oysters	Meat	Liquor	Meats			Liquor			Entire sample		
			Solids	Ash	Salt	Solids	Ash	Salt	Solids	Ash	Salt
	Per cent	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
New England:											
Unwashed—											
Maximum [1]	95.7(20)	24.4(26)	24.05(20)	3.03(15)	1.41(20)	6.67(20)	2.76(15)	2.66(20)	21.58(20)	2.70(15)	1.53(20)
Minimum	75.6	4.3	16.94	1.84	.67	4.60	2.40	1.81	15.55	1.97	.80
Average	85.07	14.93	20.57	2.29	.92	5.32	2.57	2.13	18.29	2.25	1.10
Washed—											
Maximum [1]	98.5(15)	13.9(15)	21.44(15)	1.99(15)	.63(15)	7.00(15)	1.72(9)	1.56(15)	21.0(15)	1.77(9)	.68(15)
Minimum	86.1	1.5	15.29	1.20	.24	3.62	1.08	.49	14.41	1.25	.27
Average	93.1	6.9	18.20	1.50	.35	5.01	1.34	.97	17.28	1.44	.40
New York:											
Unwashed—											
Maximum [1]			23.4(15)	2.90(15)	1.01(15)						
Minimum			17.1	1.73	.44						
Average			20.0	2.19	.73						
Washed—											
Maximum [1]			19.2(15)	1.72(15)	.34(15)						
Minimum			13.6	.98	.09						
Average			17.3	1.29	.16						
Delaware Bay:											
Unwashed—											
Maximum [1]			23.6(14)	2.10(7)	.67(14)	5.06(3)	2.18(3)	1.62(3)			
Minimum			20.2	1.74	.09	3.93	1.92	.89			
Average			22.4	1.92	.44	4.46	2.07	1.32			
Washed—											
Maximum [1]			18.69(3)		.16(3)						
Minimum			18.34		.09						
Average			18.52		.12						
Chesapeake Bay:											
Unwashed—											
Maximum [1]	97.0(22)	40.0(22)	26.9(35)	2.25(35)	.74(32)	6.65(34)	2.00(34)	1.52(34)			
Minimum	60.0	3.0	17.0	.93	.02	2.90	.64	.34			
Average	80.9	19.1	19.3	1.36	.20	4.03	1.22	.89			
Washed—											
Maximum [1]	100.0(27)	10.0(27)	17.5(27)	1.35(13)	.28(27)	5.96(18)	1.31(12)	1.05(18)	16.47(9)		.13(9)
Minimum	90.0	.0	10.73	.76	.00	2.38	.64	.18	9.98		.03
Average	95.4	4.6	14.18	1.01	.08	3.84	.78	.40	13.00		.06
South Atlantic and Florida:											
Unwashed—											
Maximum [1]	86.6(5)	47.5(5)	18.33(5)	2.72(5)	1.14(5)	4.01(5)	2.58(5)	2.05(5)			
Minimum	52.5	13.4	14.28	.80	.05	2.56	.59	.38			
Average	74.6	25.4	15.70	1.38	.36	3.20	1.20	.93			

Washed—											
Maximum [1]	95.0(7)	29.9(7)	17.47(7)	1.72(7)	.47(7)	4.57(7	1.62(7)	1.26(7)			
Minimum	70.1	5.0	13.53	.73	.0	2.14	.37	.27			
Average	87.1	12.9	14.88	1.08	.16	3.23	.86	.60			
Olympia:											
Unwashed—											
Maximum [1]	78.0(14)	43.8(14)	24.46(14)	2.74(14)	.60(14)	4.41(5)	2.72(14)	2.15(14)	20.56(6)	2.63(6)	1.47(6)
Minimum	56.2	22.0	20.10	1.43	.17	3.91	1.67	1.27	11.94	1.46	.59
Average	64.7	35.3	22.84	1.90	.35	4.14	2.32	1.79	15.02	2.40	1.22
Eastern oysters transplanted to Pacific coast:											
Unwashed—											
Maximum [1]	93.3(5)	45.6(5)	22.26(5)	2.42(5)	.82(5)	5.00(5)	2.59(5)	2.29(5)	20.56(5)	2.43(5)	1.33(5)
Minimum	54.4	6.7	19.77	1.60	.51	3.92	2.22	1.80	13.46	1.46	.59
Average	74.0	26.0	20.80	2.05	.64	4.57	2.43	2.07	16.59	1.96	.99

[1] The number of determinations from which the maximum, minimum, and average figures in each case are taken is given in parentheses after the maximum figure.

The solids content of northern oysters is higher than that of southern oysters. The moisture content of unwashed oyster meats varies between about 73 and 85 per cent, making them from three-fourths to four-fifths water. These maximum and minimum figures may not be absolutely the highest or lowest which might be found.

Oysters often contain appreciable quantities of heavy metals. Many investigators have reached the conclusion that all oysters contain some copper. According to Hiltner and Wichmann (*44*), zinc is present universally in oysters, at least in those grown in Atlantic waters. These investigators state that there is no direct relation between the zinc content and the body weight of oysters, no uniformity of ratio of zinc to copper, and no correlation between the zinc content of oysters and the water in which they grow. The quantities of heavy metals present seem to indicate that oysters exert a selective action for zinc and copper. The high proportions of zinc and copper in oysters from beds in the vicinity of industrial plants using these metals can be readily accounted for. The high zinc content of those from beds far removed from any known source of metallic contamination may be explained by the probability that oysters gradually remove traces of the metals from the water and store them in their tissues.

The arsenic, copper, zinc, and lead contents of oysters taken from beds, mostly in the vicinity of New York and New England, were determined. (Table 5.)

TABLE 5.—*Heavy metal content of oysters of known origin*

Source	Date examined	Arsenic (As_2O_3)	Copper (Cu)	Zinc (Zn)	Lead (Pb)
	1917	*Mgm. per kilo*	*Mgm. per kilo*	*Mgm. per kilo*	
Connecticut:					
Mianus River	Jan. 18	1.5	30	2,419	
		1.4	4	1,859	
Mouth of Mianus River	Jan. 30	2.2	180	1,936	
New York:					
Princess Bay	Feb. 9	1.5	117	1,453	Trace.
	Feb. 21	3.0	245	2,022	Do.
	Feb. 19	1.6	202	1,357	Do.
	Mar. 2	1.4	159	1,367	
Greenport, Long Island	Feb. 9	1.6	67	886	Do.
Oyster Bay, Long Island	Feb. 13	.6	136	1,274	
	Mar. 9	.8	196	1,734	
Great South Bay	Feb. 14	.8	98	1,107	
	Mar. 20	2.2	18	886	Do.
	Mar. 27	1.8	19	605	Do.
Oyster Bay, Cold Spring Harbor	Feb. 14	1.0	71	1,430	
East Rockaway	...do...	1.2	47	810	Do.
Rockaway	...do...	1.0	47	1,846	Do.
Hempstead Bay	Feb. 15	.6	41	1,066	
	Mar. 20	1.6	45	1,222	
New Jersey:					
Perth Amboy	Feb. 21	3.5	2,118	3,107	Do.
Virginia:					
Wachapreague	Mar. 13	2.4	12	394	

All the samples contained appreciable quantities of arsenic, copper, and zinc, and some showed traces of lead. The zinc content was very high. It was easy to trace the metallic contamination in some of these samples, for instance those from Mianus River, Conn., and Princess Bay, N. Y. On the other hand, such areas as Oyster Bay, Hempstead Bay, and Great South Bay are far from any known

source of metallic contamination, and Wachapreague, Va., is far from any manufacturing center. Yet oysters from these waters show the presence of heavy metals. There is reason to believe that oysters will absorb from the water almost any substance which it contains. Thus, oysters taken from the vicinity of dye works where quantities of aniline dyes were being discharged had distributed through their meat a variety of aniline colors.

Chemical analysis shows that oyster meat contains elements which make it a valuable food. The Lancet (*1*) reported that oysters contain all classes of nutritive material in readily assimilable form. The Office of Experiment Stations, Department of Agriculture, stated that there are 23.5 calories in 1 pound of oysters (*13*).

From its high content of soluble proteins and its high mineral salt content there is every reason to believe that the oyster is a highly valuable addition to the diet. One gallon of good-grade oysters, if not excessively washed, weighs about 8 pounds and 11 ounces. This will furnish about 1.75 pounds of dry solids, a large proportion of which is protein and valuable mineral matter. Mitchell states that the place of oysters in the dietary is not just the same as that of meat or fish, but is more like that of a cereal or vegetable. Laboratory experiments have shown that as much as 40 per cent of the solid matter in ground oyster meats is soluble in water. These water-soluble solids include portions of all the valuable food constituents of the oyster, both organic and mineral. Calcium, phosphorus, iron, and iodine are also present in oysters in appreciable quantities.

The presence of vitamins, or growth-accessory elements, in oysters has been reported by several investigators. Randoin (*73*) reported the results of such a study in 1923, and D. B. Jones, of the Bureau of Chemistry and Soils, three years later published a paper on the value of oysters from the standpoint of nutrition (*52*).

Even though a pound of oysters is nearly four-fifths water there is no waste of bone, cartilage, or other inedible part, as in meat. Oysters produced under proper sanitary conditions and not soaked excessively during washing and subsequent handling are wholesome and nutritious.

SHELLS

The results of analyses of oyster shells made by the Bureau of Chemistry are given in Table 6.

TABLE 6.—*Composition of oyster shells*

Sample No.	Constituents															
	Al	Ca	Cu	Fe	Mg	Mn	P_2O_5	SiO_2	Zn	Organic matter [1]	Water [2]	Cl	CO_2	Fl	N	As
	P. ct.	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*	*P. ct.*
1	0.045	38.78	-----	0.11	0.183	0.009	0.075	0.570	-----	1.41	0.27	0.0034	[3]57.19	-----	0.196	-----
2	.043	38.81	0.0025	.09	.189	.009	.073	.580	0.0009	1.51	.28	.0035	-----	-----	.196	-----

[1] Loss above 110° C. Ignited. [2] Loss to 110° C. [3] Average for samples 1 and 2.

Nitrogen is actually reported twice, once as the element and again as part of the organic matter. Disregarding its value as given in Table 6, 98.72 per cent of the sample is accounted for. The other 1.28 per cent may be accounted for as sodium, oxygen, and sulphates, traces of which are present in the shells.

As oyster shells are composed mainly of calcium carbonate (about 97 per cent), their principal use has been as a source of lime. This rather high grade of lime is used in agriculture as a soil improver, as a dryer for fertilizers, and for all other technical purposes for which lime is ordinarily employed. The crushed shell sometimes goes into poultry feed. Locally, the shells are sometimes used for road building. The most important use for oyster shells, however, is for cultch, to which the oyster spat may attach itself when "setting."

PHYSICAL AND CHEMICAL EXAMINATION OF OYSTERS

From the standpoint of regulatory control, physical and chemical examinations of oysters are important. For instance, if the oysters are bloated, almost white and bleached, with soft spongy texture and little flavor and with the meats almost devoid of saline taste, and if, on draining, they show much thin watery liquor, they are unquestionably adulterated with excessive quantities of water. This may have been added through soaking or floating in fresh water before or after shucking, through improper draining after washing, or, less often, through deliberate addition of water to the cans at the time of packing. The conclusion that such oysters are adulterated is further confirmed if, on examination at the laboratory, the percentage of free liquor which can be drained off is greatly in excess of 5 per cent by weight. Further, if the solids and salt contents of the meats and liquor are low, adulteration with excessive quantities of water is proved.

Thus the most important determinations in detecting watering of oysters are the physical examination and the determination of the quantity of free liquor, together with the determination of total solids and salt of the meats and liquor.

In making a physical examination of oysters, size, color, texture, flavor, and salinity (by taste) of the meats are noted. The quantity and character of the liquid present, with special note of its color, consistency, and taste are recorded.

The following determinations are almost always made when conducting chemical examinations of shucked oysters: Net volume of the contents of the can, quantity of free liquor removed by draining, percentage of meat, percentage of liquor, solids content of the meat, solids content of the liquor, chlorides in the meat, and chlorides in the liquor. The following determinations are occasionally made: Ash of the meat, ash of the liquor, protein, ether extract (fat), acidity of the liquor, and heavy metals. The following determinations are rarely made: Reducing sugars, glycogen, and composition of the ash.

Net volume of can (meat and liquor).—During shipment or storage oysters tend to pack into a solid mass and so occupy less space than when first packed for shipment. In order to loosen the mass, so that it will occupy its original volume, the oysters are poured back and forth into the standard measure before the final volume is noted.

This "fluffing" process brings them back to approximately their original volume.

Free liquor content.—The free liquor content is of very great importance as indicating probable adulteration with water. Where laboratory facilities are to be had, the determination is made with great precision, the results being reported as per cent by weight, by the following procedure:

Empty the contents of the can (if 1 gallon or larger) into a suitable receptacle, which will admit of thorough mixing. Weigh accurately on a torsion balance (accurate to 0.1 ounce) into a counterpoised pan about 1 quart (approximately 2¼ pounds) of the mixed sample. Throw this weighed sample on to a colander (preferably flat-bottomed) and drain for two minutes, receiving the liquor in a counterpoised pan. Weigh the liquor and calculate its percentage by weight. This sample can be reserved for chemical analysis.

When it is necessary to make this determination in the field, where facilities do not always permit accurate weight determinations, report percentages by volume of liquor drained off, using 1 gallon of the sample. If a standard gallon pot with vertical sides is available, this can be done conveniently by measuring the depth to the material. From this calculate the percentage of volume of liquor drained off. This gives approximate results only.

Obtaining and preparing samples for chemical analysis.—To obtain a representative sample, a quart (never less than a pint) of oysters is used. The portion used for the determination of free liquor is suitable. If the free liquor content has been found to be less than 5 per cent, the liquor may be remixed with the oysters and the whole sample analyzed. If more than 5 per cent of liquor is present, it is best to analyze the meat and liquor separately and to calculate the results to the basis of the entire sample.

Grind the meat in a food chopper, passing it through the mill twice, thoroughly mixing between each grinding. Preserve the ground sample in a tightly closed jar on ice until the examination is completed. Portions for analysis should be weighed at once. When the liquor is to be analyzed a clear solution can be obtained by placing the liquor in a tall beaker in the refrigerator. (Owing to its thick, viscous character, filtration is practically impossible.) A scum soon rises. This can be removed with a bent spatula and the clear liquor can be poured off from heavy shell particles and other materials which settle to the bottom. It is then ready for analysis.

Total solids.—Duplicate determinations should always be made. Weigh accurately 10 grams of the ground sample or of the clarified liquor into a flat-bottom platinum or nickel dish, 3 to 3½ inches in diameter. Spread the sample evenly over the bottom of the dish, using a little water if necessary to get an even distribution. Evaporate to dryness on a steam bath and dry at the temperature of boiling water to constant weight. Avoid long heating. Oyster solids easily char, and heating should be discontinued before this point is reached. Express results as percentage of solids on the meat and liquor.

Ash.—If a platinum dish has been used for the determination of the solids, this may be used for the determination of the ash. Otherwise, weigh out a fresh sample in either platinum or porcelain. Ash

the material at a low temperature in a muffle. The temperature should be kept below dull redness, as part of the ash may be lost by volatilization at higher temperatures. Oyster solids burn readily to a gray ash, free from carbon, if the temperature is not allowed to rise to a point where any fusion which occludes carbon particles takes place. Weigh the ash and report percentage by weight.

Chlorides.—Chlorides are ordinarily determined in both the oyster liquor and the meat after incineration. Direct ashing results in loss of chlorine. This may be prevented by the addition of a substance which will insure an alkaline ash. Sodium carbonate has been employed, but, on account of its fusibility, it is not entirely suitable. Calcium acetate is to be preferred.

Mix 10 grams of the sample in a platinum dish with 10 cubic centimeters of a 10 per cent solution of calcium acetate and evaporate the mixture to dryness. Then ash, preferably in a muffle at a temperature not exceeding dull redness. Take up the resulting gray ash in water acidified with dilute nitric acid (1+4). Chlorides are determined according to the official volumetric method (*7*).

Protein.—Using 2 grams of the sample, determine total organic and ammoniacal nitrogen according to the Kjeldahl or Gunning method (*7*). The nitrogen multiplied by the factor 6.25 gives the protein.

Ether extract.—Dry 10 grams of the material and transfer to a suitable continuous extractor. Extract for 12 hours with absolute ether. Regrind the sample, and continue the extraction for four hours longer. Evaporate the ether, dry the ether extract, and weigh. Report as percentage of ether extract.

Glycogen and sugar.—In routine examinations of oysters for regulatory purposes, glycogen and sugar have not generally been determined. Mitchell (*62*) and Dill (*31*) made many determinations of sugar and glycogen in shellfish. The following method, used by Dill on clams, is recommended:

Extract 25 grams of the ground sample with 85 per cent alcohol. Heat to boiling and decant through a folded paper. Then reextract with three portions of 100 cubic centimeters each of 65 per cent alcohol, heating every time. Evaporate the combined filtrates to a volume of about 400 cubic centimeters. Then follow the method of the Association of Official Agricultural Chemists (*7*).

Acidity of liquor.—Titrate 25 cubic centimeters of clarified liquor with 0.1 normal alkali, using phenolphthalein as an indicator. Express results as cubic centimeters of 0.1 normal alkali per 100 cubic centimeters of liquor.

Heavy metals.—Copper, lead, zinc, and arsenic are rarely determined in routine regulatory work. The official methods (*7*) for determining metals in food have been successful, 20 to 50 grams of ground sample being used, and the organic matter being destroyed as outlined.

Composition of ash.—The composition of ash is rarely determined. The official methods for plants (*7*), however, are applicable for the determination of the ordinary constituents of the ash of oysters.

From the regulatory standpoint, mere chemical examination of a sample of oysters means little unless something about the locality in which it originated is known. It is necessary to compare the analysis

of the unknown sample with that of an authentic sample from the same locality prepared under good commercial practices. For instance, it would be useless to compare the results of an analysis of a sample of oysters from the brackish water of Chesapeake Bay, the solids content of which, after washing, is generally between 12 and 16 per cent, with the results of an analysis of oysters from New England waters, where the solids content is generally near 20 per cent, and then attempt to draw conclusions regarding adulteration with water. On the other hand, if a type sample is available for analysis and comparison, it is easy to state whether or not the given sample is adulterated with water.

If no knowledge of the oysters' origin is available, it is best to rely upon an objective examination by one who is familiar with oysters, supported by such information as may be obtained from the simple determinations of free liquor content and solids and salt on the meat, the liquor, and the entire sample.

Spoilage in oysters generally takes the form of souring. Even before there is any pronounced odor of putrefaction there is a pronounced development of lactic acid in the liquor and in the meat. For this reason, if the question of spoilage is raised, it is best to determine acidity of the liquor. In the fresh oysters this is equivalent to only 1 or 2 cubic centimeters of 0.1 normal alkali per 100 cubic centimeters, but it rapidly increases as spoilage sets in.

Most oysters which enter interstate commerce are transported in nonreturnable friction-top cans. The gallon size is the one most commonly used, although other sizes, both larger and smaller, from 1 pint up to 10 gallons are also employed. Competition brought into service the so-called "neat" can. When commercially full this can generally holds from 1 to 4 per cent less than its supposed capacity. The difference in size between such cans and cans of the proper size is not noticeable to the casual observer. To control effectively the use of such containers and to prevent any violation of the net weight amendment to the Federal food and drugs act, a careful determination of the net contents of the can must always be made when a shipment of oysters is inspected.

The canning of oysters received attention from the Bureau of Chemistry. Cans of each size must contain a specified weight of oyster meats, and the net weight of each package must be conspicuously marked upon the label. Efforts have also been made to put an end to the practice of using for canning sour or decomposed oysters that have become unfit for the fresh-oyster market. Slack filling and using decomposed oysters for canning have been largely corrected by legal action and by educational campaigns.

OYSTERS AS CARRIERS OF INFECTION

For over a century oysters have been regarded with suspicion as potential and sometimes actual carriers of disease-producing bacteria, especially those that cause typhoid fever, dysentery, cholera, and other gastroenteric disorders.

The brackish waters of bays and streams, where oysters find the best conditions for growth and reproduction, have often been grossly polluted with sewage and trade wastes from large cities and industrial centers. At one time it was common practice to hold oysters in

storage pits and floats near the shucking houses, to allow them to "drink" in water less saline than that in which they had grown, in order to "fatten" them or give them a plump appearance, and to store surplus stocks of shell oysters until they were needed for shucking. Many cases of typhoid fever and gastroenteritis, in both the United States and Europe, have been attributed to the eating of oysters thus floated or stored in polluted water. Sometimes the epidemiological evidence seemed to establish beyond doubt that the oysters were the carriers of the infection. This conclusion was reached in spite of the fact that in almost every case it was impossible to isolate the specific organism of typhoid fever from oysters of the same locality. In other cases the epidemiological evidence has strongly indicated that oysters caused the trouble. The data, however, did not prove the case against the oysters beyond a reasonable doubt. It is not within the scope of this bulletin to discuss in detail the epidemics of typhoid fever alleged, and sometimes proved, to have been caused by the consumption of raw oysters. This is fully covered in the literature (*2*, *8*, *14*, *17*, *18*, *20*, *22*, *25*, *29*, *32*, *41*, *59*, *60*, *61*, *64*, *66*, *68*, *83*, *85*).

Very soon after the passage of the Federal food and drugs act in 1906 the combined efforts of Federal, State, and municipal agencies, with assistance from the industry, were directed toward correcting insanitary conditions in the preparation of oysters for the market. After a short period of indecision and unrest, practices in the industry were revolutionized to the extent that oysters were no longer permitted to be marketed from, or to be floated in, polluted water. Consequently, from 1916 to 1924 there were no reports of illness from infected oysters. The outbreak of a large number of typhoid-fever cases in Chicago, Ill., New York, N. Y., Washington, D. C., and 10 other cities in the eastern part of the United States during the winter of 1924–25 again focused attention upon oysters as possible carriers of disease-producing bacteria. The investigations by Lumsden and his associates (*60*), by Bundesen (*22*), and by Harris (*41*) indicated that oysters were the carriers of the typhoid-fever organism. Although the evidence against the oysters was purely epidemiological, it was so conclusive that the case appeared to be complete. The actual source of contamination could not be determined. The investigations and conferences following the report of this epidemic disclosed certain practices in producing and marketing which still needed improvement. At a meeting of oystermen and health officials at Washington, D. C., in 1925, resolutions to correct these conditions (*60*) were adopted.

Although typhoid fever is the disease most often discussed in this connection, such diseases as cholera, diarrhoea, and gastroenteritis also may be transmitted by shellfish (*19*, *24*). Two cases of illness ascribed to the eating of oysters, reported by Casey (*24*) and Brosch (*19*), are interesting in this connection. The symptoms were entirely different from those of typhoid fever; they resembled those of botulism, in that the central nervous system was affected and death was due to suffocation. In each of these cases the victim was aware that the oysters were bad as soon as he had eaten them. Botulism from shellfish is of very rare occurrence.

Obviously oysters that have been grown or floated in polluted water or that have become contaminated with filth during handling are undesirable as articles of food, regardless of whether or not they contain specific organisms of disease. The Federal food and drugs act (*91*) prohibits the shipment in interstate commerce of articles of food which are adulterated. Among other definitions of adulteration (*91*, *sec. 7*) is the one which states that a food is adulterated if it consists in whole or in part of a filthy, decomposed, or putrid animal or vegetable substance. It has been claimed by the Federal officials that oysters containing excessive numbers of *Bacillus coli* are adulterated within the meaning of the food and drugs act, and this position has been maintained through trial in court.

BACTERIOLOGICAL EXAMINATION OF OYSTERS

STANDARD METHOD

The collection of epidemiological evidence by European investigators, who were the first to study the relation of polluted shellfish to disease, was soon followed by attempts either to isolate the specific organisms of typhoid fever and cholera or to obtain bacteriological data to substantiate the claims that oysters grown in certain localities were polluted with sewage and were therefore potential sources of danger to health. The technic and culture media used by the early investigators (*21*, *42*, *46*, *51*) varied with the individual making the examination and with the investigation.

In the United States early workers on the bacteriology of the oyster also adopted methods and media varying with the individual and the ploblem (*16*, *27*, *36*, *17*, *75*, *77*, *82*).

Up to and even beyond the time when a standard method of bacteriological examination was proposed, some investigators advocated the use of the shell liquor only, some preferred the crushed meats alone, and others recommended that both the meats and the liquor be used.

The diversity of methods used in different laboratories produced confusing results that could be neither compared nor intelligently used for any well-regulated plan of sanitary control. The pressing need was for a standard method for the bacteriological examination of shellfish sponsored and approved by some organization, such as the American Public Health Association, which also devises and approves standard methods for the examination of water and milk.

Realizing this need, H. D. Pease,[3] then of the Lederle Laboratories, New York, N. Y., G. C. Whipple, of New York, and S. DeM. Gage, of the Massachusetts State Board of Health, held several conferences during 1909. As a result of these conferences, at the 1909 meeting of the American Public Health Association a committee was appointed to develop a satisfactory method for the bacteriological examination of oysters and other shellfish. The following year this committee suggested a method for the determination of total counts of bacteria and the presence of *Bacillus coli* and what is now accepted

[3] The information regarding the events immediately leading to the appointment of the Standard Methods Committee and the early history of the activities of this committee were very kindly supplied by H. D. Pease, director of the Pease Laboratories, New York, who was secretary of the original committee.

as the standard method of scoring oysters based upon the prevalence of *Bacillus coli* (*3*). In 1911 this committee modified somewhat the original procedure and considered the hibernation of oysters (*4*). In 1919 the procedure for the bacteriological examination of oysters and other shellfish now followed was approved by the American Public Health Association (*5*).

BACILLUS COLI

At various times in the development of the standard methods and the application of the results obtained it has been suggested that *Bacillus coli* scores of 23 and 32 are indicative of pollution. As further investigations have been conducted and more information has been obtained regarding the significance of this score it has become generally established among Federal, State, and municipal authorities that a *B. coli* score in excess of 50, in either shell or shucked stock, indicates excessive pollution. Any conclusion regarding the sanitary quality of the oysters based upon the bacteriological examination, however, shall be supported by data from a sanitary survey of the locality where the oysters were produced. Data obtained from a bacteriological examination can be interpreted properly only when something of the source of the oysters and the conditions under which they were handled is known.

SPECIFIC ORGANISMS

The isolation from oysters of the organism causing typhoid fever and the bacteria that are the specific causes of other diseases is impracticable. Attempts to isolate the typhoid bacillus and the cholera vibrio from oysters from polluted sources have met with but little success. Klein (*21*) was able to isolate from oysters an organism that culturally and serologically answered all the tests of *Bacillus typhosus*. From his description of the organism there is no doubt that he actually obtained the typhoid-fever bacillus. Fuller (*36*) cites an instance in which the isolation of *B. typhosus* from oysters was reported at a meeting of physicians at Constantinople. Johnstone (*51*) states that on one occasion he was able to isolate from mussels an organism which he considered to be *B. typhosus*. From his description of the isolated organism there is some doubt as to its proper identification. Finally, Stiles (*83*) isolated from oysters floated at Inwood, N. Y., an organism that was identical culturally with *B. typhosus* and that was agglutinated in a 1 to 1,000 dilution by typhoid serum. Stiles was able to isolate this organism 21 days after the oysters had been removed from water. These four instances are the only ones recorded in the literature where investigators have been able to isolate *B. typhosus* from shellfish infected under natural conditions.

Less success has been met in attempting to isolate the cholera vibrio from shellfish. Klein (*21*), among many others, made repeated attempts to isolate this organism, but without success.

It is not difficult to understand why it is practically impossible to isolate specific organisms of disease, even with the most recently improved methods of isolation and culture. The numbers of typhoid bacilli being discharged in sewage are not constant and by the time

the sewage reaches the oyster beds the dilution has become so great that any bacilli present in the effluent have become widely distributed. It is likely also that many of them have succumbed to unfavorable environment. It is not probable that each oyster on a bed will receive the same number of typhoid bacilli, so that the chance of finding the organisms in the examination of comparatively few oysters from a polluted bed is small. Naturally, it is not practicable in routine work to examine bacteriologically enormous numbers of oysters from any one bed in an attempt to reduce this hazard of sampling. Where oysters from a particular locality have been suspected as the cause of typhoid fever the epidemiological evidence, owing to the incubation period of the disease, has been obtained two weeks or more after the infection. This further removes the possibility of finding the causative organism in oysters from the suspected region. What is true of *B. typhosus* in this respect is equally true of the other pathogenic bacteria that might be present in sewage.

Because of this practical impossibility of isolating specific organisms of disease from oysters, the examination of shellfish for sanitary quality and for potentiality as a carrier of infection has depended upon tests for the presence of *B. coli*, which is the common and constant inhabitant of the intestinal tract of man and other warm-blooded animals. This organism is constantly present in sewage and can be easily isolated and identified in routine work by the use of simple cultural tests as outlined in the standard methods for the bacteriological examination of either shellfish or water.

STREPTOCOCCI AND ANAEROBES

Although but little significance is now attached to the presence in oysters of streptococci and sporulating anaerobic bacteria, numerous attempts have been made in the past to correlate their presence in shellfish with sewage pollution. Streptococci are not readily grown on the culture media used in routine bacteriological work. The presence of these organisms in large numbers may be indicative of pollution, but the difficulty of determining the significant types of streptococci and the numbers present have led bacteriologists to abandon them as reliable indices of sewage contamination.

When many samples of oysters are examined by the standard method it frequently happens that large quantities of gas are produced in lactose broth and give presumptive tests for *Bacillus coli* which can not be confirmed by streaking on Endo medium. The organisms producing this gas will not grow on the surface of solid media and, no doubt, belong to the large group of anaerobic bacteria, such as *B. welchii* and *B. sporogenes*, which have been frequently isolated from water and soil. Most of these organisms are spore formers and are resistant to the unfavorable conditions that they encounter in nature. Their presence in shellfish lacks the significance attached to the presence of *B. coli*. It has not been proved that they are of intestinal origin only. Furthermore, in the dormant spore stage these organisms may exist for long periods in water with their vitality unimpaired, and thus be obtained from shellfish growing on areas so remote from the source of pollution that all danger from nonsporulating forms like *B. typhosus* is removed. In his

investigation of the Potomac River, Cumming (*30*) found that the numbers of spores of these lactose-fermenting anaerobes were remarkably constant in the river water. The spores were often found in the best river water in 10 cubic centimeters. Their number does not indicate the degree of pollution, as does the number of *B. coli*. These spores are also often resistant to treatment with hypochlorite. Even waters so treated will contain viable spores. Several investigators have shown that these anaerobes are commonly found in sources remote from pollution with intestinal contents. Although it is true that they occur and multiply in the intestines of warm-blooded animals, their presence in shellfish can not be accepted as a reliable index of sewage pollution. If the examiner depends entirely upon the presumptive test for *B. coli* (gas production in lactose broth) he is likely to be in error, owing to the presence of these lactose-fermenting anaerobes. However, false presumptive tests are readily excluded from consideration by the failure of the anaerobes to grow aerobically on Endo plates.

POLLUTION OF OYSTER BEDS

BACTERIAL FLORA OF UNPOLLUTED OYSTERS

In order to detect the presence of sewage and filth in oysters by bacteriological methods it is first necessary to know the bacterial flora of the normal, unpolluted oyster. The presence of *Bacillus coli* can not be used as an index of pollution until it has been definitely shown that oysters do not contain *B. coli* when taken from clean and unpolluted water.

Many investigators have reported that their results show convincingly that *Bacillus coli* is not an inhabitant of normal, unpolluted oysters (*21*, *27*, *35*, *36*, *43*, *46*). In the course of routine work conducted in the Bureau of Chemistry a very large number of samples of oysters were examined by the standard method to determine the presence of *B. coli*. In many oysters obtained from sources free from pollution no *B. coli* or other lactose-fermenting bacteria were isolated. Investigations have established the fact that the normal, unpolluted oyster does not contain *B. coli*. The standard methods for the bacteriological examination of shellfish were devised with this fact in mind. Hunter and Linden (*49*) identified the organisms isolated from decomposing oysters and confirmed the findings of previous investigators (*34*, *36*, *79*, *106*) that the bacterial flora of the oyster was composed of common water and soil organisms, including cocci, nonspore-forming, Gram-negative aerobic bacilli, spore-forming, Gram-positive aerobic and anaerobic bacilli, lactobacilli, streptococci, and yeasts.

BACTERIAL FLORA OF POLLUTED OYSTERS

Oysters from polluted sources contain, in addition to the bacteria mentioned, organisms that are indicative of sewage pollution. C. A. Fuller found in polluted oysters *Bacillus coli*, *B. cloacae*, *B. welchii*, and *B. aerogenes*, none of which was found in oysters from unpolluted sources. There is still some doubt regarding the value of *B. cloacae*, *B. welchii*, *B. aerogenes*, and other such organ-

isms as indices of sewage pollution, as their origin has not been satisfactorily determined. It is frequently stated that these bacteria are commonly obtained from the soil and from other sources not generally considered as polluted. On the other hand, these organisms occur also in the intestines of warm-blooded animals and therefore arouse suspicion when found in oysters and water.

In establishing a bacteriological standard for drinking water to be used on common carriers in interstate traffic, the advisory committee appointed by the Surgeon General of the United States Public Health Service determined in what numbers organisms of the *Bacillus coli* group might be present before the water was considered unfit for use (*99*). For the purposes of the standard established the *B. coli* group was defined as in the standard method for water analysis issued by the American Public Health Association in 1923 (*6, p. 100*) "as including all nonspore-forming bacilli which ferment lactose with gas formation and grow aërobically on standard solid media." This definition of the *B. coli* group, which does not differentiate between the various forms or types of that group, would include such organisms as *B. aerogenes* and *B. cloacae*. Whether such a ruling can be applied safely to shellfish is an open question, but the presence of these organisms in excessive numbers in oysters should call for a careful survey of the territory surrounding the oyster beds, with a view to determining their possible source and, therefore, their significance in judging the sanitary quality of the oysters.

VIABILITY OF BACILLUS TYPHOSUS AND B. COLI IN OYSTERS AND WATER

In attempting to use the presence of *Bacillus coli* in oysters as an index of recent pollution, with consequent danger to the public health, it is necessary to know something of the longevity or viability of this organism in oysters and in sea water. If *B. coli* can survive for only short periods under the conditions met outside the bodies of warm-blooded animals, the use of its presence as an indicator of pollution is greatly restricted. Under such conditions water and oysters might be grossly polluted with sewage and yet not show positive tests for the presence of *B. coli*. On the other hand, if *B. coli* will multiply or survive in natural waters or oysters for extremely long periods of time, it is possible that their presence can be detected long after all danger from the pollution has ceased. *B. coli* surviving for long periods in sea water might reach oysters on beds remote from the source of the pollution. Furthermore, in order to know whether or not the presence of *B. coli* in oysters or water remote from the source of pollution indicates that the shellfish is still potentially dangerous to health some information must be had regarding the longevity of *B. typhosus* in oysters as compared with the longevity of the colon bacillus. The collection of such information has called for investigations to determine the length of time *B. typhosus* and *B. coli* can survive in sea water and in oysters floated in infected water.

Another phase of this problem of equal importance is the longevity or viability of *Bacillus typhosus* and *B. coli* in shell and shucked oysters removed from the water and stored at various temperatures. It is extremely important to know whether oysters naturally infected with *B. typhosus* on the oyster beds offer such an unsuitable

habitat for this organism that it can not multiply but quickly perishes, thus making the infected oysters safe for consumption soon after their removal from the polluted grounds. In studying epidemics of typhoid fever alleged to be due to oysters it is helpful to know whether or not *B. typhosus* can survive in stored shellfish long enough to cause the illness.

With respect to the longevity of *Bacillus coli* in stored oysters, the use of its presence as the criterion of judgment upon which the present standard method of bacteriological examination is based will fail entirely if the colon bacilli either multiply rapidly or perish in a short time within the oyster. If oysters having a *B. coli* score of less than 50 shortly after removal from the water develop a score of 500 or more during storage in the shell at low temperatures or during shipment from the Atlantic coast to the Middle West, then the bacteriological examination at destination according to the standard method loses its value. Such oysters should not be held as grossly polluted if such conditions are true.

In view of these considerations much experimental work has been done by bacteriologists interested in the pollution of shellfish to determine the longevity of *Bacillus typhosus* and *B. coli* in oysters and sea water. In fact, investigations along this line were among the first studies made on the bacteriology of the oyster. In 1889, de Giaxa published the results of his investigations of the viability of *B. typhosus* in sea water (*39*). A paper by Krumwiede, Park, and others on the longevity of this organism in oysters appeared during 1926 (*58*). The results of the outstanding investigations of this problem are summarized in Table 7.

TABLE 7.—*Summary of investigations reported in the literature on the viability of Bacillus typhosus and Bacillus coli in oysters and water*

Investigator[1]	Date	Viability of *Bacillus typhosus* in—			Viability of *Bacillus coli* in—	
		Oysters	Unsterilized water	Sterilized water	Water	Oysters
		Days	*Days*	*Days*	*Days*	*Days*
de Giaxa (*39*)	1889		9	25		
Klein (*21*)	1894	[2] 16	21			
Foote (*34*)	1895	[3] 30	[4] 8			
Wood (*106*)	1896		60			
Houston (*46*)	1901					[5] 7
Field (*33*)	1904	[6] 9,28–42	10			
Zeit (*54*)	1901		[7] 5–8, 3	[8] 15–25		
Jordan (*54*)	1901		[9] 2½			
Russell (*54*)	1901		[10] 3			
Clark (*27*)	1905				104	[11] 17
Stiles (*85*)	1912	[12] 21				

[1] Italic numbers in parenthesis refer to "Literature cited," p. 70
[2] Oysters floated in tanks of infected water
[3] Oysters stored in the shell.
[4] Very cold water used. In warmer water there was a gradual decrease after one week. *B. typhosus* was not detected after three weeks.
[5] Shell oysters stored in a "dry" condition.
[6] Shell oysters held in cold water.
[7] *B. typhosus* held in Chicago River water, which was really dilute sewage.
[8] *B. typhosus* held in celloidin sacks suspended in Lake Michigan water.
[9] *B. typhosus* held in celloidin sacks in the Chicago Drainage Canal.
[10] *B. typhosus* held in celloidin sacks in the Illinois River.
[11] Shell oysters held out of water. Oysters were dead at end of 17 days.
[12] Shell oysters held at 39° F.

TABLE 7.—*Summary of investigations reported in the literature on the viability of Bacillus typhosus and Bacillus coli in oysters and water*—Continued

Investigator	Date	Viability of *Bacillus typhosus* in—			Viability of *Bacillus coli* in—	
		Oysters	Unsterilized water	Sterilized water	Water	Oysters
		Days	*Days*	*Days*	*Days*	*Days*
Round (*75*)	1914	[14] 4			180	[23] 100
		[15] 8				
Tonney and White (*86*)	1925	[16] 22				
		[17] 8				
		[18] 60				
Jordan (*53*)	1925	[19] 24				
Kluyoun (*56*)	1925	[20] 15				
Krumwiede and Park (*58*)	1926	[21] 49				
		[22] 51				
		[23] 14–40				

[13] Shell oysters stored at 1.6° C. (34.7° F.)
[14] Shucked oysters stored at 98° F.
[15] Shucked oysters stored at 70° F.
[16] Shucked oysters stored at 45° F.
[17] Shell oysters stored at 70° F.
[18] Shell oysters stored at 45° F.
[19] Shell oysters stored at 5° C. to 8° C. (41° F. to 46.4° F.)
[20] Shell oysters stored on ice at 2.8° C. to 14.4° C. (37° F. to 57.9° F.)
[21] Shell oysters at refrigerator temperature. Oysters dead by forty-first day.
[22] Shell oysters in water cold enough to inhibit drinking.
[23] Organisms on outside of shell. Survival varied with extent of contamination.

Klein found that the cholera vibrio could be recovered from oysters on the ninth day and from sea water on the fourteenth day after infection. Stiles (*81*) stated that *Bacillus typhosus* will survive in the soil for at least 106 days. Vasquez-Colet (*100*) found that *B. typhosus* might survive on fruits and other foods eaten raw from a few minutes to three days after infection.

It seems to be well established that *Bacillus typhosus* will exist longer in sterilized sea water than in unsterilized water. This is due to the antagonism for the typhoid bacillus exerted by the other soil and water organisms present in unsterilized sea water. It is also accepted that *B. typhosus* will live longer in clean water than in water containing a large proportion of organic matter. This is illustrated by the work of Jordan, Russell, and Zeit (*54*), who found that *B. typhosus* perished quickly in the dilute sewage of the Chicago Drainage Canal but survived in the water of Lake Michigan. In view of the influence of such factors as the presence of antagonistic bacteria, the presence of organic matter, temperature, salinity, variation in resistance of individual strains of *B. typhosus*, and the number of bacilli used for the original infection, it is not surprising that each investigation gives a different result. In fact, it is impossible to draw a general conclusion regarding the viability of these intestinal organisms in water or oysters from any one set of experiments. The preponderance of the data indicates that *B. typhosus* will survive in water long enough to contaminate oysters bathed in the sewage-polluted water.

Furthermore, *Bacillus typhosus* will remain viable within the bodies or shell liquor of oysters long enough to cause illness when the oysters are eaten within the usual period elapsing between the time of removal from the infected water and the time of consumption. Many of the factors considered in the discussion on the viability of the

organism in water are significant in considering oysters. The temperature of storage, the orginal degree of infection, the condition of the oysters, and the presence of antagonistic bacteria are important. Many of the results in Table 7 are inconclusive in that the typhoid bacillus was isolated up to and on a certain date, when the experiments were terminated and no further effort was made to repeat them by prolonging the storage period. Therefore, many of the results should actually be stated as "at least" so many days or hours. In storage experiments most investigators found that during the first 24 hours there was a rapid decrease in the number of typhoid bacilli, after which the decrease became gradual, with a persistent survival of a few organisms.

The experiments reported give no reason to believe that *Bacillus coli* is any less resistant in water and oysters than *B. typhosus*. Round (*75*) stated that *B. coli* will remain viable in water for at least 180 days and that his investigations showed no multiplication of *B. coli* in oysters in cold storage at 34.7° F. Furthermore, it was present in nearly the original quantity after 100 days. Houston (*46*) found that *B. coli* persisted for more than 7 days, and Clark (*27*) stated that he recovered *B. coli* from oysters stored for 17 days, which was as long as the oysters lived under the conditions obtaining. Parsons (personal communication) states that *B. coli* will not develop in shell oysters during shipment in such a way as to affect the bacterial score. Experiments conducted by the Bureau of Chemistry showed that when shucked oysters are stored at temperatures below 50° F. no multiplication of *B. coli* or other lactose-fermenting bacteria occurs. From this observation it would seem that if oysters are stored or shipped at temperatures below 50° F., as they should be, the *B. coli* score at the end of a period of time sufficient for their transportation to market will not be greater than that originally determined at the source. However, investigations reported by Tonney and White (*87*) showed that *B. coli* did increase in shucked oysters stored at 41° to 46° F. For this reason they recommend a score somewhat higher than 50 for oysters marketed in the Middle West. Their observations are in marked contrast to those made by the Bureau of Chemistry. The optimum temperature for the development of *B. coli* is very much above 41° or 46° F. In oysters stored at these temperatures organisms which find best conditions for growth at temperatures below the optimum for *B. coli* will multiply and produce changes, making the oysters undesirable for the market before there is sufficient change in the *B. coli* score to condemn them as unfit for food.

Several conclusions can be drawn from the information at hand regarding the viability of *Bacillus typhosus* and *B. coli* in oysters and water. Regardless of the actual period of viability, which can not be determined for all conditions, the typhoid bacillus can survive in sea water long enough to infect oysters on the growing grounds and can then remain viable within the oyster long enough to cause illness. The presence of *B. coli* is a reliable index of pollution, as it also will survive in water for some time, thus contaminating growing oysters, and will remain viable for some time within the oyster if kept under proper conditions.

EFFECT OF COOKING ON BACTERIAL CONTENT OF OYSTERS

Comparatively little has been reported on the effect produced upon the bacterial content of oysters by cooking. Clark's experiments (*27*) demonstrated that the ordinary preparation of oysters for stew and pan roast does not always kill all the *B. coli* and streptococci present. In his experiments oysters were infected with *B. coli* and streptococci by floating in sea water to which sewage had been added. The oysters, which contained *B. coli* and streptococci in large numbers, were then stewed, fried, and escalloped. In certain experiments the oysters were added to milk cold and brought to the boiling point. All *B. coli* and streptococci were killed by the time the milk boiled. When oysters were added to boiling milk it was necessary to continue the boiling for at least five minutes before all the bacteria were killed. *B. coli* and streptococci were usually killed when the oysters were fried for two minutes, but they sometimes resisted frying for eight minutes. The escalloped oysters were always sterile after they had been cooked for from 15 to 30 minutes at oven temperature.

The Report of the Boston City Health Department for 1906 (*74*) states that when prepared and served as stews and pan roasts at hotels and restaurants oysters are not usually sterile. The additional heat developed between the time of cooking and serving may help to sterilize the oyster. Regardless of the effect of cooking upon the bacteria, it can not be considered a desirable practice to eat oysters from sources known to be polluted.

The effect on bacterial content produced by heating oysters at temperatures between 122° and 158° F. was studied in the Bureau of Chemistry. These experiments were not planned to demonstrate the effect of cooking oysters as practiced in the kitchen, but to determine whether or not it would be feasible to attempt to Pasteurize shucked oysters as milk is Pasteurized. It was soon evident that heating oysters at 122° F. destroyed their value for sale as anything but a cooked oyster. The conclusion is, as would be expected, that Pasteurization of shucked oysters by heat is entirely impracticable if the product is to be marketed raw.

This experiment demonstrated also that even a temperature of 158° F. applied for 20 minutes to oysters in brine will not destroy all such organisms as the staphylococci and *Bacillus aerogenes*. At 122° F. there is no appreciable effect upon the nature of the bacterial flora. If such organisms as *B. cloacae* and *B. aerogenes* survive there is little hope that the treatment would be effective in destroying *B. typhosus* or other disease-producing bacteria.

From the information at hand it is apparent that no reliance can be placed upon the ordinary cooking processes to sterilize oysters and render those which might possibly contain pathogenic bacteria safe for consumption.

RESTRICTED OYSTER-PRODUCING AREAS

In order to determine the fitness or unfitness of any particular area for shellfish production it is necessary to make a sanitary survey of this area and of the surrounding country from which drainage and pollution may reach the beds. For the purpose of ascertaining

whether or not oysters on these beds are polluted it is usually sufficient to examine the bivalves and the water in which they are grown for the presence of *Bacillus coli* and to make an inspection of the sewer outfalls and the surface pollution in the terrain immediately surrounding the area under examination. The expression "sanitary survey" in its broad sense, however, means something more than the mere examination of oysters and water for the presence of intestinal bacteria. If a general survey is to be made of a large body of water, such as a large river or bay of the Atlantic coast into which the drainage of a great many cities, towns, and smaller communities is received, it is necessary to include studies of the methods of sewage disposal in these communities, their water supply, their industrial wastes that may reach the river or bay, the prevalence of typhoid fever there, the population of the towns contributing sewage to the river or bay, and the size of the area draining into the body of water under examination. The laboratory tests made in such a survey include bacteriological examination of water, sewage, and shellfish for total numbers of bacteria and for the presence of *B. coli* and other intestinal bacteria. Chemical examinations of the water are also made to determine the oxygen demand of the sewage and the presence of nitrates, nitrites, and ammonia. The field survey and the laboratory work are supplemented by plankton studies to ascertain the numbers and kinds of protozoa and protophyta present and by hydrographic studies to show the extent to which tides affect and currents carry sewage to the oyster beds, as well as to learn the dilution of the river or bay water by fresh-water streams or by salt water from the bay or ocean.

In connection with its regulatory work on oysters from the sanitary standpoint, the Bureau of Chemistry frequently surveyed oyster-growing areas. For the purpose of gaining the information needed to recommend that certain locations be restricted for use as oyster-producing areas, it was usually sufficient to examine the shellfish and water for the presence of *B. coli* and to locate and study the immediate sources of pollution from sewers and privies. A great number of such surveys were conducted by the Bureau of Chemistry, and the results were applied in determining which areas should be restricted. The United States Public Health Service has also conducted such surveys, one of the most complete of the reports on the sanitary surveys of rivers and bays being that on the Potomac watershed by H. S. Cumming and his associates (*30*).

The pollution of rivers, bays, and harbors along the Atlantic coast with sewage and trade wastes has caused the abandonment of vast areas formerly used for shellfish production. Such excellent propagating and growing grounds as certain sections of Narragansett Bay, in Rhode Island, New Haven Harbor, Bridgeport Harbor, and South Norwalk Harbor, in Connecticut, and Jamaica Bay, in New York, have been made unfit for shellfish production by the sewage poured into them daily from the large cities and towns near by. For some years this destruction of oyster-growing areas proceeded at such a pace that the situation became very serious. In Rhode Island alone the acreage rented for shellfish growing decreased from 21,236 acres in 1912 to 9,250 acres in 1918. This decrease, 11,986 acres, resulted in a loss of revenue to that State of $70,375 a year. This shellfish

ground was abandoned partly for the reason that no "set" of oysters could be obtained on it and partly because it was so polluted that it was no longer safe as a growing ground. The failure to obtain a "set" was, in turn, held to be due to pollution with oil and other trade wastes, although this was not definitely proved.

What happened in Rhode Island happened also in other States along the seaboard. If oysters were not to become rare or extinct, especially in the North Atlantic States, it was necessary to take steps to improve conditions or to find some way of proceeding under the existing conditions. This stimulated research in the purification of oysters by floating in clean water (p. 30) and by chemical treatment (p. 33) and also in artificial hatching under controlled conditions. Although the results of these researches have been fairly satisfactory, much valuable oyster-growing ground has been lost, never to be recovered. Through the efforts of river-pollution boards and commissions organized in the various States much was done to improve conditions, but the list of areas still restricted for use as oyster-producing ground is long. There is yet room for much improvement in the sanitary condition of the bays and streams adjacent to cities and towns along the coast.

Realizing the possible danger to health and the undesirability in any event of marketing oysters from areas subject to contamination with sewage, certain polluted creeks, rivers, harbors, and bays, or portions of them have been designated by various State agencies as restricted areas for the production of shellfish. The areas restricted in each State have been decided upon as a result of sanitary surveys conducted by State officials in cooperation with the Public Health Service and the Bureau of Chemistry.

Owing to the fact that from year to year areas may be changed from restricted to nonrestricted areas, or vice versa, it is not feasible to present here a list of those now restricted. Areas which are now restricted may become nonrestricted upon the installation of properly operated sewage-treatment plants or upon improvements in the methods of handling sewage in near-by towns and cities. On the other hand, areas which are at present nonrestricted may become restricted if sewage-treatment plants, properly operated now, fail to maintain the necessary degree of efficiency or if an increase in population in the adjacent regions contributes additional sewage to the areas used for oyster production. It becomes necessary for State agencies, assisted by Federal officials, to make frequent surveys and, as a result of these surveys, to confirm the list of restricted areas or to alter it as the findings may justify.

In some States the removal of oysters for food purposes from certain restricted areas is absolutely forbidden. In others oysters may be taken from polluted areas to be transplanted into clean water where they must remain for a definite period to cleanse themselves by removing all traces of pollution. In many States it is permissible to use polluted areas for the production of seed oysters only. In at least one State certain areas are restricted for the production of shellfish throughout the entire year, while other areas, which are less seriously polluted, are restricted only during the season when the oysters are actively feeding. This ruling takes into consideration the phenomenon of hibernation (p. 26). Although no definite rules

governing restricted areas apply in every State, most oyster-producing States have adopted measures to prevent the marketing of shellfish from polluted regions.

Information on the areas restricted in any particular State may be obtained from the State board of health, the State conservation commission, the State fish and game commission, or whatever State agency may have the work in charge.

HIBERNATION OF OYSTERS

When low winter temperatures prevail in the water surrounding the beds on which they are grown oysters enter a state of hibernation. While in this condition their physiological processes are greatly retarded and, owing to a cessation of feeding, no bacteria are taken into their bodies. According to various investigators, the bacteria already within the body of the oyster when hibernation begins are rapidly digested and destroyed. Bacteriologists engaged in the routine examination of large numbers of samples throughout the year have noticed a seasonal variation in the bacterial content of oysters from the same beds. Bacteriological examination of the water over the growing areas showed that the number of *Bacillus coli* viable during the winter was the same as that during the summer, yet the *Bacillus coli* score of the oysters was very much smaller during the cold winter months than during the warm summer months. Gorham (*40*), in 1910 and 1912, and Pease (*69*), in 1911, accounted for this condition by a theory that oysters hibernated when the temperature of the surrounding water fell below a certain critical temperature.

According to Parsons (unpublished report), hibernation of oysters is not exactly the same as hibernation of nonmigratory terrestrial animals, in that hibernation of the oyster is brought about solely by low temperature, whereas hibernation of the higher animals is a natural function taking place at certain seasons, even though the temperature may remain rather high. Hibernation is a means whereby certain nonmigratory animals may survive through periods when their food supply fails and when, if normally active, they might otherwise starve. Regardless of the food supply, however, the oyster closes its shell tight and enters a period of rest or physiological inactivity when the temperature of the water falls below the critical point (45° F., according to Parsons). If a period of warm weather ensues after hibernation has begun, the oyster may be stimulated to feed again, ceasing when the temperature next falls.

Recent observations, however, have led to the conclusion that not all the oysters in an area will immediately enter into a state of hibernation or will remain in hibernation when the temperature falls below 45° F. Usually some on the bed will continue to feed, even at low temperatures. This has cast doubt upon the conclusion that hibernation is a natural function of the oyster and can be relied upon in adopting sanitary control measures.

During the warm season, while actively feeding, the oyster keeps its shell open as long as it is undisturbed. Opening and closing the shell are controlled by the strong adductor muscle attached to each valve or shell. Relaxation of this muscle in warm weather allows the shell to remain open in order that currents of water bearing

food may enter between the valves. During the active feeding period the minute organisms in the water, such as diatoms and algae, which constitute the natural food, as well as the organic matter and bacteria present as a result of pollution, are carried into the gills of the oyster, where some of this material is retained in the mucus covering the body. Cilia on the gills propel the material entangled in the mucus to the mouth of the oyster. The rapidity with which the cilia function is, to a great extent, controlled by temperature. Cold weather retards their motion so that during the hibernating season practically no food is carried in. That brings about the condition of hibernation.

It can not be claimed, however, that the shells of oysters remain tightly closed during the whole period that the temperature of the water is below 45° F. To satisfy their demand for oxygen, even in this physiologically inactive condition, the oysters must occasionally open their shells. When this occurs some food passes into the cavity, but investigation has demonstrated that the quantity is very small.

Albert Mann, of the Bureau of Plant Industry, United States Department of Agriculture, found that the stomach contents of hibernating oysters are practically destitute of diatoms. He concluded that either the oysters were physiologically inactive or the supply of diatomaceous food was very low. Parsons further demonstrated, by the use of dyes in the surrounding water, that oysters only occasionally open their shells during hibernation for the purpose of obtaining a supply of oxygen.

It is in order now to return to the rather startling results of the bacteriological examinations of oysters which first focused attention upon this so-called hibernation. In the investigation reported by Gorham in 1912 (*40*) two oyster beds in the Providence River, R. I., and two beds in the Warren River, R. I., were studied. On one bed from the Providence River oysters collected in February, when the temperature of the water was 33.8° F., scored 0, and those collected in May, when the temperature was 59° F., scored 500. At the time of both examinations *Bacillus coli* was present in 0.0001 cubic centimeter of the water. Oysters collected on a bed in the Warren River in February, when the temperature of the water was 32° F., scored 0, although the water over this bed contained *B. coli* in 1-cubic-centimeter quantities; and those collected in late April, when the water temperature was 52.7° F., scored 500, *B. coli* being found in dilutions of 1 to 100 of the water.

In 1916 Parsons and Farrar (unpublished report) found that oysters collected during November and December from grossly polluted areas in Jamaica Bay, Raritan Bay, and New Haven Harbor scored below 50, although such oysters examined during the nonhibernating period scored 500 or more. Owing to their proximity to the sewer, some of the oysters with low scores were covered with a deposit of filthy, foul-smelling mud. Obviously such oysters could not be marketed, regardless of the low scores.

In order to determine whether hibernating oysters would become polluted when floated in sewage-contaminated water, Parsons placed hibernating oysters from Raritan Bay, scoring 1, in Lupatcong Creek, which was polluted from the sewage disposal plant at Keyport, N. J. These oysters remained for six days in water at 32° to 34° F., con-

taining *B. coli* in 0.1 cubic centimeter, and sometimes 0.01 cubic centimeter, quantities, without an increase in score.

In order to show the contrast between the score of hibernating and that of nonhibernating oysters, Parsons prepared a summary of the results of his examinations of oysters from various beds conducted throughout the year. (Table 8.) The contrast between winter and summer scores would be greater had not a large number of the samples been collected at the beginning and end of hibernation.

TABLE 8.—*Effect of hibernation on Bacillus coli score*

Bed	Number of samples examined	Score with water 45° F. or lower	Number of samples examined	Score with water above 45° F.
Jamaica Bay, N. Y.	84	9	82	250
Raritan Bay, N. Y.	14	18	10	260
New Haven Harbor, Conn.[1]	3	3	6	350
Hampton Bar, Va.[1]	7	1	12	140

[1] Restricted bed.

The lowest and highest scores found in oysters from certain locations, with the date and the temperature of the water at the time the samples were collected, are given in Table 9. These results show that the low scores of winter are not due to any diminution of the number of colon bacilli in the water over the beds, the variation in the *B. coli* count of the water being only that due to tides, currents, wind, and unusual pollution from land sources as a result of heavy rains.

TABLE 9.—*Seasonal variation in Bacillus coli scores of oysters from polluted areas*[1]

Area	Minimum score			Maximum score		
	Score	Temperature of water when found	Date when found	Score	Temperature of water when found	Date when found
		°F.			°F.	
Jamaica Bay, N. Y.:						
Big Channel	2	44	Mar. 31, 1914	500	72	July 14, 1915
Island Channel	4	32	Jan. 21, 1914	500	72	May 14, 1915
Fishkill Channel	1	31.5	Jan. 5, 1915	500	72	Do.
Pumpkin Patch	0	31.5	....do....	500	67	Sept. 16, 1914
Ruffle Bar	1	31.5	....do....	500	72	May 14, 1915
Beach Channel	2	30.5	Jan. 23, 1914	500	70	Sept. 8, 1914
Silver Hole	1	46	Apr. 15, 1914	500	64	Sept. 24, 1915
Grass Hassock	0	30	Jan. 23, 1914	410	62	May 18, 1914
Broad Channel	2	42	Nov. 20, 1914	140	70	Sept. 6, 1914
Irish Channel	5	31.5	Jan. 5, 1915	320	64	May 25, 1914
Horse Channel	5	32.5	Jan. 26, 1914	140	64.5	June 1, 1914
Old Swale Channel	3	34	Mar. 17, 1914	140	64	Do.
Raritan Bay, N. Y.:						
Bed 998	5	38	Jan. 28, 1915	500	76	July 15, 1915
Bed 989	23	43	Nov. 17, 1914	410	70	Aug. 10, 1914
Bed 1031	0	38	Jan. 28, 1915	230	76	Do.
Bed 995	4	34	Jan. 18, 1915	320	69	June 24, 1914
Beds 985 and 980	0	38	Jan. 28, 1915	410	75	July 15, 1915
Bed 996	4	40	Dec. 22, 1914	410	76	Aug. 10, 1914
Bed 891	3	38	Jan. 28, 1915	320	70	Do.
New Haven Harbor, Conn.: City Point	2	44	Nov. 27, 1916	500	52	Nov. 5, 1915
Hampton Bar, Va.	0	32	Feb. 12, 1917	410	53	Nov. 15, 1916

[1] Compiled from unpublished report of P. B. Parsons, 1917.

From the information at hand it is evident that low temperatures of the sea water surrounding the oyster beds bring about a resting or dormant condition in the oyster. This cessation of feeding, in turn, produces a condition within the oyster approaching sterility, the bacterial content being reduced to almost zero.

Oysters on beds in the waters of New York, Connecticut, Rhode Island, and Massachusetts usually go into hibernation during the latter part of November and remain until late in April. The oysters in lower New Jersey hibernate a little later and resume normal activity a little earlier in the spring. The oysters in Maryland and Virginia may not go into hibernation until a month later and the period of hibernation is often comparatively short.

The fact that during hibernation the bacterial content of oysters and the possible menace to health from the consumption of shellfish are reduced to a minimum has been applied by some States to regulations for sanitary control. Certain areas on the border line between a grossly polluted and a clean condition are utilized for oyster production between December and March, when the temperature of the water remains below 45° F.

It must not be assumed that because hibernating oysters are bacteriologically clean within the shell the same is true of the outside of the shell. Deposits of mud and filth collect on the shells of oysters growing near sources of pollution. Upon shucking, this mud and filth find their way into the shucked product to constitute a real menace to health. The number of intestinal bacteria in the water is no smaller during the cold weather than during the warmer seasons, and the outside of the shell may be grossly contaminated with bacteria that menace the health and with other material that offends the sense of decency, even when the oyster within the shell is clean.

Although some States permit the use of oysters, during hibernation, from slightly polluted grounds, other States, in spite of the well-established fact of hibernation, do not allow oysters to be taken for sale from areas during the winter unless they are fit for use during the warmer seasons. Recognizing the fact that the water over the beds is no cleaner in the winter than in the summer and that the mud and filth on the shells will contaminate the oysters during shucking, the State of New York condemns for oyster production throughout the year all areas that are unfit during the nonhibernating seasons.

Data on hibernation prove that oysters, at least those in northern waters, enter into a state of physiological inactivity when the temperature falls to 40° to 45° F. During this period of inactivity the bacterial content of the oyster is reduced to a minimum. Whether this phenomenon should be taken advantage of in any system of sanitary control is a question which has not been satisfactorily agreed upon by all State and Federal agencies. Even admitting the facts regarding hibernation as here set forth, it is probably safer and wiser to prohibit the use of oysters from areas at any time of the year unless those areas are fit for production during all times of the year.

PURIFICATION OF OYSTERS

TRANSPLANTING

When transplanted to clean water, free from contamination with sewage, actively feeding oysters that have become polluted on the growing areas cleanse themselves within a reasonably short period. The length of time depends primarily on the purity of the water. While the oysters are feeding actively the gills act as a filter to strain out some of the material that may be brought in by the 7 or 8 gallons of sea water which pass through each oyster daily. If this sea water contains sewage with large numbers of intestinal bacteria, some of the microorganisms are entrapped in the mucus on the body of the oyster and are then transferred to the alimentary tract by the currents of water set in motion through the action of the cilia on the gills. When oysters thus polluted are floated in clean sea water containing no intestinal bacteria, or very few, the microorganisms already present are eliminated and no more are ingested. In this way the oyster purifies itself in a remarkably short time.

The rapidity of this cleansing process under varying conditions has been investigated by many bacteriologists. For several reasons it is important to know the minimum time in which a proper cleansing can be effected after transplantation. Injury may result to oysters held too long under unnatural conditions. If the oysters are floated in water of lower density than that in which they were grown, bloating will take place. If prolonged, this results in their death. The undesirability of using for floating purposes, over long periods, areas that may be needed for growing oysters is apparent. In case of a demand for shipments on comparatively short notice the grower or dealer must know the minimum length of time necessary for the purification of any polluted oysters he may be obliged to use. Therefore, the important consideration in the investigations undertaken has been to determine beyond doubt the time necessary to effect a proper cleansing without prolonging the period to the point where it becomes injurious to the oysters or expensive to the growers. Regardless of expense and inconvenience, however, the primary object of such transplanting must be to rid the oyster of all traces of sewage pollution.

In Europe, Klein (*21*), Herdman and Boyce (*42*), and Johnstone (*51*), and in the United States, Phelps (*70*) and Round (*75*) have investigated the ability of oysters to cleanse themselves.

In Table 10 are presented the data obtained in the investigations on the effect of transplanting polluted oysters to unpolluted water conducted by Parsons in Connecticut, New Jersey, and Virginia. These indicate that 24 hours suffices to cleanse polluted oysters if the water to which they are transplanted is really clean and if the temperature, the salinity of the water, and the season of the year are conducive to active feeding. Such conditions are usually found with difficulty, as ordinarily the creeks or bays used for transplanting are subject to occasional pollution during the floating period and the salinity of the water of the creeks may change greatly with the change of tide, becoming practically fresh at the end of the ebb tide. The effects of occasional pollution and of changing salinity are seen in the results of the floating experiments in New York State, at Hassock Creek,

Bigg's Creek, John's Creek, Garretson's Creek, Winant's Pond, and Flatlands. Here the reduction in score was greatly retarded. It is apparent that repeated cleansing with occasional contamination ultimately brings about a purification of the oyster, although it is somewhat delayed.

TABLE 10.—*Results of transplanting polluted oysters in unpolluted water*[1]

Bed	Date	Time floated	Temperature of water	Salinity of water	Score	
					Beginning of period	End of period
		Days	*°F.*	*Sp. gr.*		
East Haven River	October, 1916	1	55-56	1.019	320	14
		1	55-56	1.019	320	32
		2	55-57	1.019	3,200	14
Buckroe Beach, Va	November, 1916	1	50-51	1.021	1,400	4
Old Point Comfort, Va	do	3	50-52	1.021	500	41
Hassock Creek [2]	September, 1914	14	60	1.022	500	32
	do	3	72	1.022	230	32
	May, 1915	7	61	1.021	320	5
Bigg's Creek [2]	October, 1914	4	49	1.022	320	23
John's Creek [2]	do	4	49	1.022	320	41
Garretson's Creek [2]	do	4	49	1.022	320	23
Winant's Pond [2]	do	4	60	1.018	140	23
Flatlands [2]	do	3	60	1.020	140	23

[1] Compiled from data submitted by P. B. Parsons to Bureau of Chemistry, 1917.
[2] The water to which the oysters were transplanted was subject to occasional contamination during the floating period.
[3] The water to which the oysters were transplanted was fairly clean, but became fresh at the end of the ebb tide.

The effect of occasional pollution during the floating period was very well illustrated by experiments conducted by Farrar (unpublished report) at Block Island, R. I. Polluted oysters were transplanted to New Harbor, which is ordinarily free from pollution. At the time of the experiments the waste from some warships moored near by polluted the water. Consequently, there was no reduction in the *Bacillus coli* score during the first 48 hours. The ships then departed and during the following 24 hours a purification of the oysters was accomplished. Farrar's experiments demonstrate the possibility of cleansing highly polluted oysters within 48 hours. Oysters were artificially infected by floating near a sewer for several days until scores of 50,000 and 140,000 were obtained. Floating for 48 hours in clean water reduced the scores to 5 and 4. This was a severe test, as oysters do not have such high scores naturally. Oysters grown so near the sewer would be smothered by deposits of mud and filth and would be rendered unfit for market.

In experiments conducted in 1918 at Wickford, R. I., Hunter found that the score of oysters when transplanted to clean water was reduced from 5,000 to 14 and from 410 to 32 in 5 days. In another experiment 96 hours was sufficient to reduce scores of 410 and 230 to 14 and 23. At certain times the water of Wickford Harbor contained *B. coli* in 1-cubic-centimeter quantities and at other times no *B. coli* could be found in the 10-cubic-centimeter quantities. From this it is evident that the water of Wickford Harbor was not uniformly clean but was subject to periodic contamination. This accounts for the fact that from four to six days were required for the oysters to cleanse themselves.

Although it is true that polluted oysters will cleanse themselves when transplanted to clean water, it is equally true that clean, unpolluted oysters will rapidly become polluted if floated in sewage-contaminated water. Data taken from a report submitted by Parsons in 1917 showing to what extent this pollution of oysters occurs are presented in Table 11. When *Bacillus coli* was present in 0.01 cubic centimeter of the water of New Haven Harbor, Conn., it required only two hours to increase the score of the oysters floated there from 14 to 500. Hibernating oysters floated in Lupatcong Creek for seven days did not increase in score. This fact, together with the data given by Round (*75*) showing that the score of hibernating oysters did not decrease when transplanted to clean water, lends further support to the theory of hibernation or physiological inactivity of oysters at low temperatures.

TABLE 11.—*Results of transplanting unpolluted oysters in polluted brackish water*[1]

Bed	Date	Time floated	Temperature of water	Salinity of water	Score	
					Beginning of period	End of period
		Days	*°F.*	*Sp. gr.*		
East Rockaway Creek, N. Y.	October, 1914	1	60	1.013	3	23
					5	50
Lupatcong Creek, N. J.	June, 1915	5	70	(2)	32	140
					23	500
New Haven Harbor,[3] Conn.	October, 1916	½	56	1.019	3	41
New Haven Harbor,[4] Conn.	November, 1916	½	56	1.019	14	500
Lupatcong Creek,[5] N. J.	March, 1915	7	40	(2)	(6)	5

[1] Compiled from data submitted by P. B. Parsons to Bureau of Chemistry, 1917.
[2] The water in this creek had a salinity of 1.017 at the end of the flood tide and was nearly fresh water at the end of the ebb tide.
[3] *B. coli* were present in 0.1 cubic centimeter of the water of the harbor where floating took place.
[4] *B. coli* were present in 0.01 cubic centimeter of the water of the harbor where floating took place.
[5] These oysters were in a hibernating condition.
[6] Not given.

From all the investigations conducted it is apparent that polluted oysters can purify themselves when transplanted to unpolluted waters. Although the length of time necessary for such purification can not be determined definitely for all locations from the results of the studies already made, it can be concluded that such purification is possible within 24 hours if the conditions are exactly right and that seven days will suffice, even when the water to which the oysters are transplanted is subject to occasional pollution. Much discretion must be exercised in applying to sanitary control the knowledge that oysters have the power of self-purification. Before the merits of any location can be passed upon, study must be made of the water and attention must be given to the sanitary features of the surrounding country. It is suggested that transplanted oysters remain in the clean water for about seven days. If the water is clean, as it should be, and if the conditions are such that the oysters feed normally in freely circulating water, this period will allow a margin of safety over the time absolutely necessary for the cleansing. This margin of safety is necessary because of the impossibility of controlling all the factors involved in the process. Whenever pos-

sible, *Bacillus coli* scores should be obtained on the transplanted oysters before they are taken up for the market, and no oysters should be taken for sale until the score has been reduced below 50. As oyster samples from one location sometimes vary, there is danger that the results of one examination may be misleading. Therefore, the low score should be obtained on two or more samples before the oysters are considered fit for use.

If the water in which the oysters are transplanted is clean, if the currents and the arrangement of the oysters are such that there is free circulation of pure water among them, and if the temperature is favorable for active feeding and for discharge of polluting material from the shellfish, this ability of oysters to cleanse themselves can be used to help solve the problem of how to utilize stock from polluted areas that have been condemned.

CHLORINATION

The lack of natural clean waters, suitable for the purification of oysters by floating and within a reasonable distance from the polluted areas, made it necessary to search for some other practical means of purification. For instance, pollution made it necessary either to abandon Raritan Bay, N. Y., an excellent growing area, or to find some means of artificial purification, as no near-by waters could be used for self-purification. This need for a method of artificial purification, which existed also in other localities, led to the development of the chlorination process by Wells, with the assistance of representatives of the United States Bureau of Chemistry, the United States Public Health Service, the oyster industry, and the New York State Conservation Commission (*23*, *101*, *102*, *103*, *104*, *105*).

The biological principle underlying this method is the same as that upon which self-purification is based. In fact, the two methods are the same except that, in the method developed by Wells, by the use of chlorine as a sterilizing agent, clean water is provided for floating. The ability of the oyster to cleanse itself by washing out impurities when floated in clean water is made use of in this method under carefully controlled conditions.

Wells, experimenting in 1914 and 1915 at Fisherman's Island, Va., found that if oysters were floated in water containing an excess of free chlorine they would close their shells, permitting sterilization of the exterior, and that when the excess of free chlorine was greatly diminished they would open their shells again, thereafter cleansing themselves in the sterilized sea water. In 1916 this chlorination process was tested, with the assistance of Parsons of the Bureau of Chemistry, at New Haven, Conn. In 1920 the Public Health Service, the Bureau of Chemistry, and the New York State Conservation Commission cooperated to demonstrate the process under commercial conditions. This demonstration, which was followed by an indorsement of the method and certification by the New York State Conservation Commission of a purification plant at Inwood, Long Island, is reported in detail by Carmelia (*23*). Polluted oysters in large lots were spread in great wooden floats and covered with clean sea water. Calcium hypochlorite, in such quantity that free chlorine was available in from 4 to 6 parts per million, was distributed over

the floats by means of hand-operated wooden paddles. The quantity of chlorine needed was determined by the quantity of organic matter in the water. The greater the quantity of organic matter the greater is the need for chlorine. More chlorine can be used than is tolerable in drinking water. From 20 to 30 minutes after the addition of the hypochlorite the water was tested with orthotolidine for free chlorine. The oysters were then left undisturbed during a "drinking" period of 6 hours. At the end of this period the floats were again treated with chlorine as at first, after which they were left for from 12 to 18 hours. This gave a 24-hour "drinking" period, at the end of which the oysters were ready for the market.

The hypochlorite first sterilized the water in which the oysters were placed. For a short time this excessive quantity of chlorine was so irritating to the oysters that they repeatedly and forcibly ejected water from between their shells. This action removed much mud, sand, and other organic matter. The action of the chlorine also loosened the organic matter on the exterior of the shells. Left undisturbed, the oysters filtered the sterilized water through their gills and alimentary tracts, removing materials which served to moderately pollute the floating water. The second treatment with calcium hypochlorite again sterilized the water, in which the oysters were then floated for 18 hours.

In these experiments a reduction of 90 per cent of the *Bacillus coli* content of oysters scoring 50 or more was obtained. According to Carmelia (*23*), a reduction of 90 per cent of the *Bacillus coli* in oysters scoring 500 will produce a score of 50, bringing the oysters only to the passing mark and allowing no margin for safety (Table 12). He believed, therefore, that no oysters which scored higher than 230 or 320 should be purified by this method.

TABLE 12.—*Bacteriological results of oyster purification by hypochlorite process*[1]

Source	Run No.	Quantity of oysters treated	Average *Bacillus coli* score		Average reduction of *Bacillus coli* score
			Before treatment	After treatment	
		Bushels			*Per cent*
Raritan Bay between Great Kills and Princess Bay, N. Y.	1	46	23	2	91.3
Do	2	50	50	5	90.0
Jamaica Bay, Big Channel, Carnarsie, N. Y.	3	20	230	23	90.0
Raritan Bay between Great Kills and Princess Bay, N. Y.	4	60	14	4	71.4
Do	5	150	14	4	71.4
Do	6	60	5	3	40.0
Do	7	50	4	2	50.0
Do	8	60	14	5	64.3
Raritan Bay, off South Beach, N. Y.	9	15	41	4	90.2
Raritan Bay between Great Kills and Princess Bay, N. Y.	10	60	2	1	50.0
Raritan Bay near Great Beds, Light Amboy, N. Y.	11	25	320	4	98.7
Jamaica Bay, Sweet Water area, near Inwood, N. Y.	12	20	320	3	99.0
Jamaica Bay, Big Channel, Carnarsie, N. Y.	13	15	320	23	92.8

[1] Reprinted from Carmelia's report (*23*).

As a result of the indorsement by Federal and State authorities of the chlorination process as an efficient and practical method of cleansing oysters, a plant for this purpose was installed on a commercial basis at Inwood, N. Y. (*101, 102, 104*). At present the State of New

York does not permit the taking of oysters from beds where *B. coli* scores of over 50 are obtained, except in the summer for the purpose of transplanting. This regulation was put into effect as a safety measure, but its enactment did away with the need for a chlorinating plant such as the one at Inwood, which has been dismantled. However, several oyster companies at West Sayville, N. Y., are now using the chlorinating method upon most, if not all, of the oysters handled by them.

In its control of the chlorination process the New York State Conservation Commission has issued the following regulation (*67*):

> Raw water to be purified for use in drinking, floating, or the water storage of oysters shall have an initial score of not more than 5 (5 portions of the water sample to be treated as 5 shellfish in securing this score). Said water shall have a salinity sufficient to give a specific gravity of not less than 1.007 at 15° C. and shall be treated with liquid chlorine, calcium or sodium hypochlorite, or similar chlorine compounds produced by electrolysis. If the fill and draw method is used, sufficient chlorine or its compounds in one of the forms mentioned shall be added to the water to secure a free chlorine content of not less than 0.5 parts per million in any part of the tank fifteen minutes after filling is completed. If the continuous flow method is used, a control reaction chamber shall be provided through which the water shall be passed before it enters the oyster storage basin. Such chamber shall be twice as long as it is wide and not less than 4 feet deep, properly baffled and having a detention period of not less than fifteen minutes. In using this method, sufficient chlorine or its compounds in one of the forms mentioned shall be added to the water to give a free chlorine content of not less than 0.5 parts per million to the water collected from the outlet of such reaction tank before it enters the tank in which the oysters are stored.

The chlorination method is efficient if carefully supervised and properly conducted. It should be as carefully supervised as is the Pasteurization of milk. Perhaps more important than the quality of the oysters used, the purity of the water, the strength of the sterilization agent, or the length of the "drinking" period, are the intelligence and care shown by the operator. It is absolutely necessary that the operator have enough experience to understand the principles underlying the process and to be able to interpret his results as they are obtained.

The experiments of Krumwiede, Park, and others (*57*) indicate that, under the experimental conditions, the chlorination treatment is not reliable in ridding badly contaminated oysters of viable typhoid bacilli. They conclude that "the chlorination treatment of contaminated oysters will result in a marked diminution in the number of *B. typhosus*, but even six successive treatments may not rid the oysters of the contaminating pathogens. The process can not be recommended therefore in any sense as a reliable means of 'sterilizing' contaminated oysters and thus rendering them safe for consumption." The experiments upon which this conclusion was based represent extreme conditions, in that typhoid feces containing enormous numbers of bacilli and large quantities of organic matter were added to relatively small volumes of water in which oysters were floated. This was followed by the use of such quantities of chlorine that it is possible that the normal functions of the oyster in purifying itself were interfered with. There is still plenty of reason to believe that oysters contaminated under natural conditions on beds not too grossly polluted with sewage may be purified by the chlorination process. It has not been recommended that attempts be

made to chlorinate and to purify for use as food grossly polluted oysters.

At some shucking houses where surplus stock is stored in tanks or floats until needed, it is considered good practice to add calcium hypochlorite to the water in which the oysters are stored. This is done to prevent contamination between the time the oysters are taken from the growing areas and the time of shucking.

At first thought, the chlorination of oysters may not appear desirable. It is suggestive of an attempt to render fit, by chemical disinfection, a filthy and polluted product naturally unfit for use. Such objections to chlorinated oysters, however, are based on a lack of understanding of the process. It is becoming as difficult to produce oysters entirely free from pollution as it is to find sources of drinking water that is naturally fit for use. In making drinking water clean and safe mechanical filters and chlorine are used. There is no objection to this in the minds of the users of such water. Similar use of chlorine is made in the artificial purification of oysters. Furthermore, to quote Wells (*103*), "this method of purification consists of nothing more than assuring conditions of cleanliness under which the oyster can, by its natural function, remove any pollution received from the water."

The treatment with chlorine produces no change in the appearance of the oyster nor in its flavor. The calcium hypochlorite reacts with the salts present in the sea water to form calcium carbonate and sodium chloride, both of which are normally present in sea water. It is no doubt true that, in oysters scoring not too high originally, the finished product after chlorination is cleaner bacteriologically than some of the oysters marketed direct from the growing areas. It may be that sooner or later the opinion of the advocates of this process will prevail and that chlorination of oysters will be regarded in the same light as chemical treatment of drinking water and Pasteurization of milk. At present sufficient investigational work has been conducted to demonstrate that, when properly carried out, the chlorination method is not objectionable and offers a means of rendering clean, safe, and fit for market, oysters which otherwise could not be used.

As its efficacy depends upon the power of oysters to cleanse themselves by "drinking" the clean water in which they are floated, it is obvious that this method can not be applied to shucked oysters. However, some dealers are adding chlorine to the water with which the oysters are washed in the shucking house. The proponents of this method of cleansing shucked oysters claim that about 10 parts per million of free chlorine, available in the wash water, prevents any increases in the *Bacillus coli* score and in the total numbers of bacteria during washing and handling subsequent to shucking.

FLOATING OYSTERS IN THE SHELL

For a great many years before the passage of the Federal food and drugs act it had been known that floating living shell oysters in brackish or fresh water would bring about great changes in the proportions of solids and water in oysters. In 1887 and 1888 Atwater (*9*, *10*, *11*, *12*) discussed the application of the principles of osmosis and dialysis and presented data to show the extent of the changes

taking place during floating. He directed attention to the fact that, as a result of floating, there was a gain in volume and a loss of solids and salt and that the appearance and flavor of the oysters were greatly changed. In spite of the knowledge that such changes, amounting to adulteration with water, were produced in oysters by floating, this practice was continued.

In the face of claims by certain oyster shippers that it was necessary to float shell oysters in order to cleanse them, to remove some of the salt that they might stand shipment better, and to meet a popular demand for a plump, "fat" oyster, it was necessary for the Bureau of Chemistry to do some investigational work to determine whether or not there was a real need for floating and to ascertain to what extent adulteration occurred when oysters were floated.

As a result of this work and of hearings held in 1909, Food Inspection Decision 110 (*88*) was issued. This decision stated, among other things, that oysters are adulterated within the meaning of the act if they have been subjected to floating or "drinking" in water containing less salt than that in which they were grown. Protests by certain oyster interests called for further hearings, as a result of which Food Inspection Decision 121 (*89*) was issued. This decision permitted the floating of oysters in water of lower salinity than that in which they would grow to maturity, if the packages in which they were shipped were labeled "Floated oysters."

At the time of the issuance of Food Inspection Decision 121 the distribution of floated oysters was not widespread and the sale was largely in the shell, or, if shucked, by count. Later the shipment of floated shell oysters for shucking and sale by volume presented another aspect of the problem. If floated oysters have greater volume than unfloated stock from the same source, the indications are that fraud will be perpetrated upon the consumer who receives an excess of water and a deficiency in oyster solids. This practice of selling floated oysters by volume stimulated further research on the chemistry of the process.

Early in the 1916 season experimental shipments of floated and unfloated oysters were made from New Jersey points to Philadelphia, Pa., and to Baltimore, Md. Examination of the stock at destination showed that the volume of the shucked floated oysters was about 25 per cent greater than that of the same stock that had not been floated. Also, the floating had removed a large part of the salt, had materially changed the flavor, and had reduced the solids content about 20 per cent. The experiment at that time indicated some apparent superiority in shipping and keeping quality in the floated oysters as compared with the unfloated oysters. This observation is interesting in view of the results obtained from experiments subsequently conducted.

In January and October, 1922, and in the early fall of 1924, further experiments were conducted. The cold season of the year was chosen for the first experiment, as it was believed that with the temperature of the water low the oysters would be inactive and, consequently, results different from those previously obtained when the floating was carried on before the temperature of the water had fallen below 40° C. might be expected. The second experiment was conducted during October, when the weather was unusually warm, so that

the oysters were subjected to practically summer temperatures during storage and handling.

In the first experiment oysters from two areas and in the second experiment oysters from three areas were examined after removal from salt water and after removal from brackish water at high and low tide, following floating periods of 42 and 46 hours in the first experiment and 36 to 42 hours in the second experiment. Several sacks of oysters were used in each experiment. The data (Table 13) are the average results on each lot examined. In the second experiment the salinity of the river in the vicinity of the floats, expressed as grams of salt per 100 cubic centimeters, ranged from about 0.7 at high tide to 0.2 at low tide. At the same time the salinity of the water over the oyster beds ranged from about 1.8 to 2.4. The unfloated and floated oysters were shucked, measured, weighed, and counted. A sample from each type was withdrawn and analyzed.

TABLE 13.—*Changes in oysters floated in the shell and removed at high tide and at low tide*

Experiment No.	Treatment	Yield per sack	Gain in volume	Weight per gallon	Count per gallon (average)	Volume of individual oyster	Solids [1]	Salt [1]
		Gallons	*Per cent*	*Ounces*		*C. c.*	*Per cent*	*Per cent*
1	Lot 1:							
	Unfloated	4.110	.....	141.76	250.0	15.2	22.60	0.56
	Floated 46 hours; removed at high tide	4.444	8.0	140.30	239.0	15.8	21.40	.52
	Floated 42 hours; removed at low tide	4.423	7.4	141.03	239.0	15.8	21.50	.48
	Lot 2:							
	Unfloated	3.254	.....	140.87	288.0	13.3	19.80	.71
	Floated 46 hours; removed at high tide	3.645	12.0	139.86	272.0	13.9	18.90	.61
	Floated 42 hours; removed at low tide	3.772	15.9	140.00	266.0	14.2	18.80	.55
2	Unfloated from deep water	3.078	.....	138.22	238.0	15.86	19.46	.89
	Floated; removed at high tide	3.801	23.6	135.94	213.7	17.70	15.63	.30
	Floated; removed at low tide	4.423	43.7	135.57	180.9	20.97	14.93	.23

[1] Entire sample.

Although the procedure followed was practically the same in both experiments, the results did not agree exactly. In the first experiment the physical examination on shucking showed the oysters of each type to be in good condition, even though some had been frozen during shipment. There was no indication that floating, even to low tide, when the temperature was very low, as in this case, had done any serious injury nor was there anything to indicate that unfloated stock would not ship as well as floated stock. In the second experiment, although all the oysters were in marketable condition, there was a marked difference in the physical appearance of the unfloated and floated stock. The floated oysters taken at low tide had lost the creamy color, the firm texture, and the excellent saline flavor of the unfloated oyster and had become pale, bleached, soft, and spongy, with an insipid and flat taste. The floated oysters removed at high tide showed physical characteristics between these two extremes, but more nearly resembling the low-tide floated oysters.

In both experiments increased yield per sack, gain in volume, increase in size of the individual oyster, loss of weight per gallon, a

reduced number of oysters per gallon, and a loss of solids and salt as a result of floating were in evidence.

In the first experiment the weight of the individual oyster increased from 0.4 to 0.8 gram, according to the tide on which it was removed. As a result of the floating there was a reduction in solids of approximately 5 per cent. In the second experiment the increase in yield produced by floating at low tide amounted to 30 or 40 per cent and about half this increase if the floating was discontinued at high tide. These results are in contrast with those obtained when the water was below the temperature where the oyster functions actively, in which case the gain produced by floating was only about 10 per cent. From these two experiments it is apparent that the changes produced by floating are greatly influenced by the temperature of the water and the consequent physiological activity of the oysters.

An experiment conducted in the early fall of 1924 included, in addition to the points covered in the previous experiments, a study of the changes which the oysters, both unfloated and floated, undergo during longer storage periods in the shell under the influence of warehouse storage at ordinary temperatures and in artificially chilled storage. The effect produced by ordinary commercial washing on the volume of shucked oysters obtained from unfloated and floated stock was also studied.

The plan of the experiment was to prepare and examine 14 lots of 3 sacks each of unfloated and floated oysters. The oysters used were taken from water having a salinity of about 1.65 grams of salt per 100 cubic centimeters and they were floated in water having about 1.2 grams of salt per 100 cubic centimeters at high tide and about 0.2 gram at low tide. The lots of oysters used were removed from the floats on the high tide or the low tide. The results of this experiment are given in Table 14.

TABLE 14.—*Average weight and count per gallon, size, and composition of unfloated and floated oysters*

Lot No.	Treatment	Weight per gallon	Count per gallon	Volume of individual oysters	Solids	Salt	Solids per gallon	Average decrease in count	Average increase in volume	Decrease in solids per gallon
		Ounces		*C. c.*	*Per cent*	*Per cent*	*Ounces*	*Per cent*	*Per cent*	*Per cent*
1	Unfloated	135.18	220	17.2	18.43	0.106	24.91			
2	do	135.35	215	17.6	18.69	.158	25.29			
3	Floated; high tide	134.83	193	19.6	16.91	.029	22.80			
4	do	135.14	202	18.8	16.71	.091	22.58	11.0	12.6	12.9
5	do	133.77	182	20.6	14.50	.055	19.40			
6	do	134.90	191	19.5	16.00	.079	21.60			
7	do	135.24	198	19.1	16.91	.114	22.87			
8	Floated; low tide	134.74	184	20.6	13.89	.027	18.72			
9	do	134.01	171	22.1	13.69	.038	18.17			
10	do	133.61	167	22.7	13.17	.020	17.47			
11	do	134.27	171	22.1	13.53	.023	18.35	20.6	25.3	26.8
12	do	135.11	174	21.8	13.57	.020	18.33			
13	do	134.45	178	21.2	14.30	.051	19.23			
14	do	134.82	169	22.4	13.49	.044	18.16			

Of those removed from the floats at high tide, lots 4 and 7 were floated 7 and 5 hours, respectively, from low tide to high tide; lot 5 was floated 12 hours, from high tide over one low tide; lot 3 was

floated 18 hours, to two high tides; and lot 6 was floated 42 hours, from low tide over two tides. Of those removed at low tide, lot 9 was floated 7 hours, from high tide to low tide; lots 8 and 13 were floated 13 and 11 hours, from low tide to low tide; lot 10 was floated 24 hours, from low tide over one low tide; lot 11 was floated 37 hours, from low tide over two tides; and lots 12 and 14 were floated 48 hours, from low tide over four low tides.

Within 40 hours after removal from the water, two sacks of each lot were shucked and examined, the third sack being reserved for examination after a storage period. At the time of shucking visual inspection showed that both the unfloated and floated oysters were in excellent marketable condition, although there was no difficulty in distinguishing the floated from the unfloated stock, owing to the pale, bleached, and bloated appearance of the former, especially of those removed from the floats at low tide. The floated oysters appeared larger than the unfloated oysters, contained much free liquor in the shell, and emitted large quantities of milky liquor when pierced.

Each lot of oysters was washed as described in Table 15. Before and after washing the oysters were measured and after washing they were also weighed, counted, and analyzed for solids and salt. Quantities of each lot were shipped in ice to Buffalo, N. Y., where they were re-iced and returned to Baltimore, Md., to be examined for free liquor.

TABLE 15.—*Changes in volume produced by commercial washing of unfloated and floated oysters*

Lot No.	Treatment of shell oysters	Treatment of shucked oysters	Gain in volume	Loss in volume
			Per cent	*Per cent*
1	Unfloated	Blown for 2 minutes with 50 gallons of water	5.9	
		Washed in tub for 2 minutes with fresh water; agitated	7.7	
2	do	Blown for 2 minutes with 50 gallons of water	7.4	
3	Floated; removed at high tide.	Washed in tub for 2 minutes with fresh water; agitated	.0	0.0
4	do	Blown for 2 minutes with 50 gallons of water		4.1
5	do	Washed in tub for 2 minutes	4.5	
6	do	Washed in tub for 2 minutes with fresh water	.0	.0
7	do	Blown for 2 minutes with 50 gallons of water	.0	.0
8	Floated; removed at low tide.	do	2.0	
9	do	Washed in tub for 2 minutes with fresh water	.0	.0
10	do	Blown for 2 minutes with 50 gallons of water		1.9
11	do	Washed in tub for 2 minutes with fresh water	3.9	
12	do	do	.0	.0
13	do	Blown for 2 minutes with 50 gallons of water	.0	.0
14	do	do	.0	.0

The changes in volume produced by the washing process are given in Table 15. The average figures obtained on the weight, count, volume, solids content, and salt content of the unfloated and floated oysters are given in Table 13, which shows also the average percentage changes in volume, count, and solids content. The remarkable feature brought out by this experiment is that the length of time of floating had practically no bearing on the changes produced, the determining factor being whether the process was terminated during a low tide or a high tide. The oysters floated for seven hours and removed at low tide showed almost identically the same gain in volume and change in composition as those floated for 48 hours and

removed at low tide. The same condition was noted in the oysters removed from the floats at high tide.

The free liquor drained off the unfloated oysters returned from Buffalo ranged from 3.7 to 2.1 per cent, an average of 2.6 per cent; that of the floated oysters removed at high tide from 8.9 to 4.7 per cent, an average of 7 per cent; and that of the floated oysters removed at low tide from 14.4 to 6.8 per cent, an average of 9.9 per cent. These figures indicate that oysters floated at low tide developed large quantities of liquor during transit—quantities larger than those normally present in properly washed and skimmed oysters after shipment.

The residual sacks of oysters were stored first for 10 days at a temperature of about 30° F. At the expiration of this period one-half of the oysters in each sack were opened and examined visually and the other half stored for 6 to 10 days longer in an ordinary shucking room exposed to prevailing temperatures. At the end of both periods the oysters were living and in marketable condition. There was still a difference between the unfloated and floated stock, but both showed evidence of drying about the gills. After washing, both types of oysters presented a fair appearance and were, in fact, marketed commercially, in spite of the fact that they had been out of the water for 20 days, half of the time exposed to warm weather. This does not indicate that floating is necessary in order to produce an oyster which will stand any reasonable shipment or storage in the shell.

These experiments show that the duration of the floating period has little effect on oysters, that floating is not necessary to produce oysters which will stand shipment well, and that floating in fresh water materially changes the composition of oysters, especially if the floating period is terminated at low tide.

As a result of this experimental work, Food Inspection Decision 211, Adulteration of oysters (*90*), was issued in June, 1927. This decision revokes Food Inspection Decision 121 (*89*) and reaffirms Food Inspection Decision 110 (*88*). The shipment of floated oysters, even if labeled "Floated oysters," now constitutes a violation of the Federal food and drugs act.

SHUCKING-HOUSE SANITATION

Clean oysters, improperly handled, in an unclean and insanitary shucking house, may become so contaminated that when shipped they are unfit for use. Federal and State authorities do what can be done to regulate and control conditions upon the growing grounds, but the responsibility for sanitary conditions in the shucking house rests squarely upon the dealer or shipper. Federal and State inspectors may assist the shipper by calling attention to conditions open to improvement from the sanitary standpoint. Such inspections, made in cooperation with the shipper rather than with any idea of hampering him or interfering with legitimate practices, can be made only occasionally. Therefore, it is the dealer himself who must assume the burden of keeping a clean house in order to deliver a clean product.

Phelps (*71*) states that the quality of oysters depends upon (1) the character of the water from which they are taken, (2) the process

of handling from the shucker to the shipping package, and (3) the treatment which they receive from this point to the consumer.

In 1911 Stiles (*82*) directed attention to the importance of sanitation within the shucking house. He stated that without the devices necessary for cleansing and sterilizing it is impossible to prepare the final shipping package in a sanitary manner. Specific suggestions for the maintenance of sanitary conditions within the shucking house were made by Round in 1916 (*76*).

Beginning with the premises outside the shucking house proper, it is suggested that orderliness and neatness be maintained. It is understood that the ordinary practices of the industry make it necessary to have shell piles, stocks of reserve materials, etc., on the premises. No objection can be made to such accumulations. Dirt, piles of decomposing materials, pools of stagnant water, and other similar nuisances should not be tolerated, however. A neat and orderly outside appearance has its effect upon visitors and passers-by, as well as a direct bearing upon the cleanliness of the product shipped.

For several reasons it is advisable to use whitewash and paint liberally within the shucking house. Painting or whitewashing should be done at least once a year, and more often if necessary. The walls and the ceiling must be freed from dirt and cobwebs before they are painted or whitewashed. Paint and whitewash are germicides and their use also gives better light and a better appearance. Thus paint and whitewash have a direct effect on the quality of the shucked oysters and an indirect effect, through the impressions made upon the employees that they are working in a clean shop and must be clean themselves.

If possible, the floors and benches should be made of concrete instead of wood. Concrete can be kept clean more easily. When bins and benches are emptied they should be cleaned from all accumulations of mud, dirt, and seaweeds. This can be done by hosing thoroughly, or, if sufficient water pressure is lacking, by scrubbing with brooms and rinsing with plenty of water. The floors should be washed daily in the same manner.

Particular care is necessary to provide and use absolutely clean utensils. Cans, colanders, knives, skimmers, and tanks should be cleaned and rinsed whenever empty or not in use. A liberal use of steam for cleaning and sterilizing them is necessary. Sufficient steam for this purpose should be provided, regardless of the temperate weather of fall and spring, which does not call for steam for heating purposes. The steam may be used as live steam or under pressure. If steam can not be provided, a solution of calcium hypochlorite may be used as a cleansing and sterilizing agent. The routine use of hypochlorite solutions for rinsing bins and benches is to be recommended because of its sterilizing and deodorizing properties.

Convenient toilets for employees should be provided. These toilets should have running water and they must be used carefully. Employees should be impressed with the fact that they are handling foods that may be eaten raw. Therefore, they must always wash their hands after using the toilet.

Doors and windows of the shucking house should be screened tightly to prevent the entrance of flies and other insects.

Much has been written regarding compulsory periodic physical examinations of all food handlers, such as cooks, waiters, and oyster shuckers. As applied to oyster shuckers such a regulation would be of value if it eliminated from the shucking house all carriers of *Bacillus typhosus* and all sufferers from venereal or other contagious diseases. In the absence of such a requirement the dealer or shipper is under an obligation to inquire carefully about the health of each employee and about any past case of typhoid fever or other contagious disease. It is also important that the supervisor in the shucking house be advised of any illness in the family of each employee. There is always danger that disease-producing organisms may be transmitted from homes where illness prevails by persons who are not themselves suffering from the disease. From the knowledge gained regarding the health of each employee and his family the employer should draw conclusions and act accordingly. Bearing in mind that oysters offer a favorable medium for the development of bacteria and are frequently eaten raw, the employer should rigidly exclude from work in the shucking house all persons suffering from contagious disease or known to be harboring organisms of disease although not suffering themselves.

Personal cleanliness, of course, must be insisted upon. The habits and person of the shucker should be clean. The shucker's hands must be kept as clean as possible in order not to unduly contaminate the shucked oysters with mud and débris in the process of opening. In spite of all precautions, it is possible that some unhealthy person or some one carrying disease organisms may be employed. Employees should be impressed with the idea that they are working in a kitchen where food is prepared.

WASHING OYSTERS

IN THE SHELL

Where oysters are grown on muddy or soft bottoms the shells may become coated with mud. Unless removed before shucking, this serves to contaminate seriously the shucked product.

Where power dredges are used and deck space is available, much of the mud, if soft and not sticky, may be removed when the oysters are first taken from the water. This may be done by rinsing them several times in the sea water while still in the dredge and just before dumping on the deck. When dumped on the deck the oysters may be washed with pailfuls of water taken from alongside the boat. When the oysters are taken by means of tongs by men working in small rowboats this washing is not so easily done. Furthermore, if the bottom soil is claylike and sticky it is removed from the shells with difficulty, even when wet, and, if allowed to dry, such shells can be cleaned only by scrubbing.

Under the present system of handling at the shucking house no method is provided for cleansing the shells before shucking. Whether serious attempts to develop such a method have been made by oyster growers is unknown, but the literature apparently contains no references to it.

It is often stated that it is impracticable to attempt to rid the exterior of the oysters of mud at the shucking house, but the impor-

tance of this mud as a factor in the contamination of the shucked stock would seem to warrant some expense and inconvenience on the part of the shipper in an attempt to eliminate it. Although it might be difficult to put such an operation into effect in some of the smaller, poorly equipped shucking establishments, the installation of suitable apparatus would not seem too difficult or expensive for the larger companies. The importance of this contaminating factor where muddy oysters are handled can not be overemphasized.

OUT OF THE SHELL

Shucking is always followed by washing. The efficiency of the various methods of washing in removing bacteria from the shucked oysters has been the subject of extensive investigations. In 1904 Houston (*46*) established by experiment the fundamental principle that oysters polluted on the growing areas can not be washed free from *Bacillus coli*. By none of the present commercial methods of washing can the original *Bacillus coli* score be reduced. The efficiency of a washing method is measured by the extent to which it removes the dirt and the bacteria introduced during the shucking operations. The bacterial content of the oysters frequently increases greatly, owing to the introduction of mud and other foreign material during shucking. Hibernating oysters and oysters from sewage-free waters scoring as low as 5 often score 500 after shucking. In 1916 and 1917, Parsons and Farrar reported the results of their experiments upon the efficiency of various methods of washing as affecting the bacterial content of oysters. The data in Table 16 show the effect of the shucking and washing processes upon the bacterial content of oysters.

TABLE 16.—*Effect on score of shucking and of different methods of washing*[1]

Method of washing	Condition of oysters	Score		
		Before shucking	Before washing	After washing
Hosing on skimmer with tap water	Hibernating	(2)	500	500
Do	do	(2)	14	23
Holding in tub of fresh water:				
15 minutes	do	4	(2)	4
30 minutes	do	14	(2)	500
20 minutes	do	2	230	320
	do	2	320	50
	do	50	410	500
	do	41	500	140
	do	0	50	41
30 minutes	do	2	32	32
	do	4	41	140
20 minutes	do	0	23	41
	do	0	32	50
30 minutes	do	2	50	50
	do	5	500	500
20 minutes	Nonhibernating	41	(2)	500
	do	32	(2)	410
	do	410	(2)	410
	do	140	(2)	4,100
	do	230	(2)	5
Hosing before and after immersion in tubs	(2)	4	50	[3] 4
Do	(2)	5	500	500

[1] Compiled from data submitted by P. B. Parsons in 1917.
[2] Not given.
[3] There were few oysters on the skimmer. Plenty of water was used.

TABLE 16.—*Effect on score of shucking and of different methods of washing*—Continued

Method of washing	Condition of oysters	Score		
		Before shucking	Before washing	After washing
Agitated in blower:				
7 minutes [1]	Hibernating	5	32	14
6¼ minutes [2]	do	5	320	32
3 minutes [1]	do	5	140	50
5 minutes [1]	do	14	230	14
5 minutes [2]	do	14	140	5
7 minutes [1]	do	0	23	4
5 minutes [1]	do	0	14	1
3 minutes [1]	do	0	14	0
5 minutes [1]	do	0	14	0
3 minutes [1]	do	0	14	0
3 minutes [1]	do	3	14	3
5 minutes [2]	do	2	230	4
10 minutes [2]	do	3	32	3
10 minutes [2]	do	2	320	3
5 minutes [1]	do	4	320	14
7 minutes [2]	do	3	230	5
5 minutes [1]	Nonhibernating	41	320	41
5 minutes [2]	do	320	1,400	230
30 minutes [2]	do	320	1,400	320
10 minutes [1]	do	320	2,300	410
30 minutes [2]	do	410	500	410
Washed in running water under tap for 30 minutes	do	23	50	50
Do	do	320	500	230

[1] Fresh water was used in the blower. [2] Salt water was used in the blower.

In many houses it is customary to wash the shucked oysters immediately after shucking on a perforated skimmer with running water from a hose. Such washing has little effect on the bacterial content. It is usually superficial. The oysters on the skimmer are so numerous that the agitation of the mass by the force of the water and by stirring with the hand or a paddle is insufficient to expose the entire surface of each oyster to the stream of running water. Many bacteria adhering to the mucus of the body and to the gills escape the action of the water and are carried on to the finished product. After ordinary hosing on a skimmer, the scores were the same as before washing. If a few oysters spread out on the skimmer are treated with plenty of water, under strong pressure, it is possible to effect a slight reduction in the score. Under commercial conditions this would greatly retard operations. As ordinarily practiced, hosing of oysters on a skimmer can not be held to be an efficient means of washing.

By another method of washing, shucked oysters are held in tubs or tanks of fresh or salt water and are occasionally stirred with a paddle. The results of experiments to demonstrate the effect on the score of holding oysters in tubs of fresh water for varying lengths of time are given in Table 16. The oysters, collected during the hibernating season, had low scores before shucking. As a result of contamination during shucking the scores were greatly increased. Holding the oysters in tubs for as long as 30 minutes did not remove the bacteria introduced during shucking and thus did not reduce the scores to those originally found. No differences in results were obtained when salt water was used in place of fresh water. Data submitted by Parsons and Farrar show that, owing to the use of dirty

water, the score may be increased during the washing process. When care was taken to use clean water for each batch of oysters and to stir the mixture frequently some slight reduction in score was effected. The indications that stirring had much effect in reducing the score were too meager to be of very great significance and subsequent work indicated that stirring has little effect on the efficiency of this process.

Further experiments showed that washing in tubs of fresh water polluted oysters taken during the actively feeding season does not reduce the score. (Table 16.) Polluted oysters washed in this manner still had scores as high as those not washed. These results indicate that it is impossible to remove *Bacillus coli* from the body of polluted oysters by washing.

Experiments conducted to determine the effect of hosing oysters before and after immersion in tubs of water (Table 16) showed that such hosing adds nothing to the efficiency of the washing process. Some of the results support the statement (p. 45) that a few oysters spread on the skimmer and treated with plenty of water can be reduced in bacterial content.

The investigations conducted by Parsons indicated that the most practical and efficient method of washing oysters to reduce the contamination introduced by shucking is the use of the blower. The blower is a metal tank equipped with a perforated plate just above the bottom. Air is forced up through the perforations in this plate into the mixture of water and oysters. By the force of the air the oysters are tossed about and agitated violently, thus exposing the entire body to the washing action of the water or brine. Dirt, pieces of shell, and slime, sinking to the bottom of the tank, are collected in the space between the bottom and the perforated plate. This accumulation of waste must be removed after each operation and clean water must be used for each blowing. Otherwise the material gathered in the bottom of the tank is redistributed throughout the wash water and the effect is to further contaminate the oysters rather than to cleanse them. The main objection to the use of the blower is that prolonged blowing may bloat the oysters and thus adulterate them with water (p. 47). The use of a weak salt solution will eliminate this trouble.

The results of experiments on clean oysters collected during the hibernating season and on polluted oysters collected during the actively feeding season (Table 16) show that by use of the blower the score of shucked oysters can be reduced to that of the oysters before shucking. No reduction in the score of the polluted oysters below that of the original shell stock could be made, however. Occasional hosing of the oysters before blowing was resorted to, but with little effect on the bacterial content. It was found that, for cleansing purposes, there was no difference between the use of fresh water and the use of weak brine in the blower. Also as good results were obtained by blowing for from three to five minutes as by blowing for a longer period.

Of all the present commercial methods of washing studied the use of the blower was the only one that was efficient in reducing the bacterial content of the shucked oysters.

Another experiment (Table 16) was conducted to learn the effect of washing oysters in tubs or buckets under rapidly running streams

of water for prolonged periods. Nonhibernating oysters were subjected to this treatment. The results were negative in so far as they show any effect upon the score. Such a washing process will not reduce the score below that of the oysters before washing.

To sum up the present knowledge of the bacteriology involved in the washing process, hosing or spraying the oysters with water on a skimmer will not reduce the bacterial contamination due to shucking and handling, but washing in a blower in a weak brine for from three to five minutes will reduce the bacterial content to about that of the oysters before shucking, without undue bloating. No known method of washing will remove from the body of the oyster evidence of pollution gained during growth on the beds.

EFFECT ON VOLUME AND COMPOSITION

The composition of shucked oysters can be greatly changed by prolonged washing in fresh water. If the process is prolonged unduly the oysters become adulterated within the meaning of the Federal food and drugs act. So much fresh water is absorbed by the oysters that bloating or "fattening" takes place and fraud is perpetrated on buyers of such oysters. Furthermore, valuable soluble solids are lost in the wash water or liquor that is drained off and discarded. Although any fresh-water washing must result in some loss of soluble solids, as well as in the addition of water, a reasonable washing of shucked oysters is necessary to obtain a product free from shell, dirt, and foreign material. The process must be regulated to a degree that will permit a thorough cleansing without soaking and adulterating.

Realizing the possibilities for illegitimate gain to dealers from the increase in volume due to soaking, and recognizing also the probability of adulteration during the washing process due to the shippers' lack of knowledge of the chemical changes taking place, the Bureau of Chemistry studied the effects produced by various methods of washing oysters.

During the season of 1916–17 oyster-washing experiments were conducted near Providence, R. I., New Haven, Conn., New York, N. Y., and the Chesapeake Bay (Md.) section (reported by E. J. Shanley). The washing process employed in the establishment under investigation was noted and the experiments were planned to simulate such processes. The volume of oysters was measured before and after washing, and the change of volume under the conditions was noted. Samples of oysters for analyses were collected before and after washing. The procedures followed in these experiments are briefly described and the results obtained are given in Table 17. Data on the solids and salt content of the meat and liquor of the samples, before and after washing, also were obtained. Only the determinations on the entire samples are given in the table.

TABLE 17.—*Summary of results of oyster-washing experiments conducted at New Haven, Conn., and Providence, R. I., 1916–17*

Locality and experiment No.	Method of washing	Meat		Liquor		Whole sample				Change from washing		Gain in volume
						Solids		Salt				
		Unwashed	Washed	Unwashed	Washed	Unwashed	Washed	Unwashed	Washed	Solids	Salt	
New Haven, Conn.:		*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
1	Sprayed with hose 1 minute; stood in tub of water 1 hour	86.48	92.57	13.52	7.43	17.86	14.41	1.05	0.56	−19.31	−46.6	(1)
Greenport, N. Y.:												
2	Washed in blower 3 minutes; sprayed on skimmer 1 minute	85.40	94.70	14.60	5.30	17.20	15.39	1.12	.48	−10.52	−57.1	(2)
3A [3]	Blown in fresh water 3 minutes; washed with running water	87.93	98.50	12.07	1.50	21.06	17.50	.87	.30	−16.90	−65.5	15
3B [3]	Blown in fresh water 3 minutes	88.70		11.30		20.97	18.77	.80	.28	−10.49	−65.0	4
Narragansett Bay, R. I.:												
4	Blown for 5 minutes, with stream of fresh water running continuously through blower.	85.60	89.60	14.40	10.40	20.40	17.51	1.02	.28	−14.17	−72.5	9
5	Washed on skimmer ½ minute; stood in tank of water, with occasional stirring, 20 minutes.	83.30	86.10	16.70	13.90	20.61	18.26	1.02	.47	−11.40	−53.9	2.5
6	Sprayed on skimmer 1 minute; blown in fresh water 5 minutes.	86.70	95.90	13.30	4.10	21.58	20.30	1.03	.35	−5.93	−66.0	12.5
7	Passed along riffle board over which water constantly flowed.	81.10	87.80	18.90	12.20	18.46	17.34	1.13	.54	−6.07	−52.2	(2)
7A	Oysters from experiment 7 collected in tank and agitated with stream of fresh water until tank overflowed; washing continued 15 or 20 minutes.	81.10	90.90	18.90	9.10	18.46	17.31	1.13	.31	−6.23	−72.6	7.5
8	Blown in fresh water 3 minutes					22.82	16.92	.82	.29	−25.85	−64.6	9.2
8A	do					22.82	18.48	.82	.35	−19.02	−57.3	8.0
8B	Blown in 1.5 per cent brine 3 minutes					22.82	20.91	.82	.66	−8.36	−19.5	(2)
New Haven, Conn.:												
9	Blown in tank through which fresh water constantly flowed 5 minutes.	87.86	94.17	12.14	5.83	16.15	13.01	1.02	.37	−19.44	−63.7	5.0
Narragansett Bay, R. I.:												
10	Blown in fresh water 3 minutes					19.97	14.14	1.04	.42	−29.19	−59.6	8.0
11	Sprayed on skimmer 1 minute; stood in weak brine 2 minutes; stirred a few times; some brine poured off; oysters again stood 5 minutes.	90.50		9.50		21.50	19.80	.82	.32	−7.90	−60.9	1.8

[1] Slight loss. [2] None. [3] 2 lots of oysters used.

Although the 11 experiments described can not be considered as accurate scientifically, in that all the factors were not controlled, they show the increases in volume and losses of solids and salt caused by various washing methods. The results indicate that (1) salt and solids are extracted from the oysters by any form of washing; (2) when oysters are washed with fresh or salt water in tubs, or with salt water in a blower, the volume increases very little; and (3) when washed with fresh water in a blower the volume increases greatly. It is evident that the tub or blower method of washing, if carried to excess, will cause adulteration. If properly controlled, however, none of these methods of washing need cause serious adulteration with added water.

As the water content of oysters varies somewhat with the region in which they are grown and as different methods of washing are practiced in different localities, experiments similar to those conducted in New England were carried out in New York and in the vicinity of Chesapeake Bay. At nine establishments near New York, in eight of which a blower was used, experiments under strictly commercial conditions were conducted with oysters from Princess Bay. The oysters were sprayed for about 1 minute on a skimmer and were then blown for periods varying from 5 to 10 minutes.

The maximum loss of solids in any one experiment was 18.8 per cent and the minimum loss was 4.5 per cent. The average loss of solids in the nine experiments was 11.8 per cent. The maximum gain in volume was 18 per cent and the minimum 4.2 per cent, or an average gain of 8.2 per cent. The salt was largely removed in the washing process.

Of other experiments conducted near New York one indicated that when 1 per cent salt solution is used in a blower the loss of solids is about one-third and the gain in volume about one-fifth that obtained when fresh water is used. Another showed that when the ratio of oysters to water in the blower is increased the gain in volume is greatly decreased.

In the Chesapeake Bay region the methods of shucking and washing differed greatly from those farther north. It is the practice in the south Atlantic industry to shuck the oysters into buckets which are one-third to one-half full of fresh water. After being passed over a skimmer the shucked oysters are collected in large tubs partially filled with fresh water. The collection of shucked oysters in these tubs is continued until 30 to 50 gallons have accumulated. This means that some of the oysters are soaked in varying quantities of fresh water for periods up to two to four hours.

In order to investigate the effect of such a process on the composition, samples were obtained before and after the washing process from a number of shucking houses. The loss in solids during washing was determined, but no figures are available to show the gain in volume, if any. The effect of this method of washing on the solids and salt content of the oysters and the percentages of solids and salt present in unwashed oysters from the Chesapeake Bay region are given in Table 18.

TABLE 18.—*Composition of Chesapeake Bay oysters before and after washing*

Locality	Method of washing	Solids	Salt	Loss in solids
		Per cent	*Per cent*	*Per cent*
Tangier Sound [1]	Unwashed	20.8	0.34	
	Washed	17.9	.14	13.9
Tangier Sound and Potomac River [1]	Unwashed	17.4	.58	
	Washed	13.7	.11	21.3
Potomac River [1]	Unwashed	17.7	.29	
	Washed	15.8	.15	10.8
Pocomoke plants [2]	Shucked in water and rinsed	17.8	.15	0.5
	Unwashed	21.4	.37	
	Shucked in water and drained.	20.1	.14	6.0
	Washed	20.3	.21	5.1
Fishing Bay [2]	Unwashed	18.7	.43	
	Shucked in water	17.8	.19	4.8
	Washed	16.0	.13	14.4

[1] Unwashed oysters shucked into dry containers. Liquor was drawn off before sampling.
[2] Washed oysters shucked into containers containing water, then collected in tubs of water and soaked for various lengths of time, depending on factory practice, or probably not longer than 2 or 3 hours.

As it had been observed that unwashed oysters "bled" or excreted liquor more freely than washed oysters, experiments were conducted to study this more extensively. The results of these experiments, based on washings in tubs and blowers, confirmed the conclusion that unwashed oysters "bleed" profusely as compared with washed stock. Furthermore, when the original liquor was drawn off and discarded the unwashed oysters continued to "bleed" and were soon surrounded with more liquor. This "bleeding" was stopped by immersing the oysters in fresh water.

This observation led to a laboratory experiment to determine how long after shucking "bleeding" would continue and to ascertain the composition of the excreted liquor. The more torn the oyster meat, the greater is the excretion of liquor. Therefore, careless opening, which cuts and tears the meat, tends to increase the quantity of liquor that will separate on shucking. The experiment consisted in (1) separating the liquor within the shell, (2) separating the liquor excreted during the shucking, (3) separating the liquor that accumulated when the shucked oysters were allowed to stand for 30 minutes, and (4) separating the residual liquor produced during the 20 hours following shucking, over and above that which had been removed within the first 30 minutes after shucking. The quantity and composition of these separated fractions are given in Table 19.

TABLE 19.—*Quantity and composition of liquor within the oyster shell and of that excreted during and after shucking*

Liquor	Proportion of total contents	Solid matter	Ash	Salt	Protein (N×6.25)
	Per cent	*Per cent*	*Per cent*	*Per cent*	*Per cent*
In shell	5.1	2.43	2.09	1.58	0.14
Excreted during shucking	12.2	3.23	1.90	1.56	.85
Excreted on standing 30 minutes	26.2	3.57	1.94	1.52	1.23
Residual, separating during 20 hours after standing 30 minutes	2.9	4.57	1.91	1.46	1.68
Separated oyster meats 30 minutes after shucking [1]		17.2	1.76	.58	

[1] Represented 56.5 per cent of the contents of shell 30 minutes after shucking.

Apparently the first liquor in the shell surrounding the oysters was largely salt water, containing little organic matter and a very small quantity of nitrogenous material. During the 30-minute standing period the meats "bled" freely, resulting in the separation of 26.2 per cent of liquor. During an additional 20-hour standing period but little further "bleeding" occurred. It is significant that the percentage of solid matter in the liquor progressively increased, until the last portion of liquor examined after standing for 20 hours had a solids content of 4.57 per cent. The nitrogenous constituents had increased also until they represented nearly half of the total solids present.

Shucked oysters as prepared for market are never transported for any distance in their own liquor. After being washed in water they are passed over a skimmer, the excess liquid is drained off, and the oysters are packed "dry" in cans. Improper draining results in adulteration. Water so embodied gradually takes up the solids until it shows a composition closely approximating that of the natural liquor found in the shell at the time of shucking. Therefore it would be extremely difficult to determine by examination of this liquid whether it was, in fact, oyster liquor or unremoved wash water unless the quantity of liquid that properly prepared oysters contained after shipment was known.

Accordingly, for the purpose of studying in a more elaborate way the effect produced on volume and composition by various methods of washing, as well as on the free liquor content produced by storage and shipment, experiments were carried out during the oyster seasons of 1921 to 1924, inclusive. These were of three types: (1) Tub washing and soaking experiments; (2) washing experiments, using a blower; and (3) experiments to determine the effect on liquor content produced by storage and shipment (p. 62). The results for types 1 and 2 are given in Table 20.

TABLE 20.—*Effect of washing oysters in tubs and with blowers.*

Source of oysters	Washing process	Gain in volume	Loss in solids	Loss in salt	Free liquor on standing
		Per cent	*Per cent*	*Per cent*	
Potomac River	2 gallons of oysters with 3 gallons of water in tub, 1½ hours.	.....	6.5	21.4	
Do	2 gallons of oysters with 3 gallons of water in tub, 3 hours.	.....	22.0	64.0	
Do	2 gallons of oysters with 3 gallons of water in tub, 4½ hours.	14.0	26.0	64.0	Appreciable quantity.
Nansemond Ridge, Chuckatuck, Va.	20 gallons of oysters with 50 gallons of water in tub, 4 hours, stirred every 15 minutes.	8.1	15.0	72.7	None at end of 18 hours.
Great Wicomico River, Md.	20 gallons of oysters with 50 gallons of water in tub, 4 hours, not stirred.	6.3	16.0	77.7	Small quantity.
Do	20 gallons of oysters with 50 gallons of water in tub, 4 hours, stirred frequently.	13.8	24.3	83.3	Do.
York River, Va	20 gallons of oysters with 50 gallons of water in tub, 5 hours, without stirring.	5.6	17.0	68.3	None.
	20 gallons of oysters blown:				
West River-Choptank, River, Md.[1]	3 minutes	None.	6.2	62.5	Do.
Do	6 minutes	None.	8.2	87.5	Do.
Potomac River	3 minutes	6.0	12.4	64.2	Do.
Do	6 minutes	7.5	16.7	80.0	Do.
Back River, Va	3 minutes	None.	6.4	60.9	Do.
Back River and Mobjack Bay, Va.	6 minutes	3.7	9.7	58.3	Do.
Maurice River, N. J.	3 minutes	7.5	18.2	59.0	Do.
Do	6 minutes	8.7	18.0	59.0	Do.

[1] These oysters were thin and of poor quality.

As in all other washing experiments, there was some gain in volume and some loss in solids. The gain in volume was influenced by the condition of the oysters. Those used in the first and third experiments with a blower were thin and did not absorb much water, whereas those in the second and fourth experiments with the blower were plump and gained greatly in volume. It was concluded from the first tub-washing experiment that the loss in solids was practically complete in three hours. The results of the third tub-washing experiment indicated that the gain in volume can be increased by agitation of the oysters in the tub. In none of the eight washing experiments was much free liquor excreted on standing.

Two experiments in which oysters were washed in tubs and two in which blowers were used were conducted in 1922 and 1923, with the object of determining (1) the change in volume and composition of oysters subjected to a reasonable commercial washing, (2) any additional change by continuing the washing process until it became in effect a soaking process, (3) the quantity of oyster solids dissolved and carried off in the wash water, and (4) how much of the water absorbed during washing is permanently retained and will not appear as free liquor after shipment.

The two tub-washing experiments (No. 5 and No. 6) were conducted along very similar lines. The only real difference between them was that, in No. 6, owing to a scarcity of stock, it was necessary to use, for the prolonged soaking period, a fresh lot of oysters on a succeeding day.

In order to simulate commercial conditions wherein oysters are allowed to collect and remain in tubs of fresh water from 2 to 4 hours, the oysters in experiment 5 were allowed to remain in tubs of fresh water for 5 hours, being examined before washing, at the end of 2¾ hours, and at the end of 5 hours. In experiment 6 they were soaked for a longer period, being examined before washing, at the end of 2 hours, and at the end of 6 hours. The oysters used in experiment 5 were of unusually inferior quality, the meats being attenuated and badly torn. Those used in experiment 6 were of better quality.

In both experiments, before washing and at the end of each of the soaking periods, various determinations were made on the oysters and the wash water. Each gallon of oysters was measured in a standard gallon pot and weighed on accurate scales to 0.01 ounce. The solids in the oysters, in the hydrant water used for washing, and in the resulting wash water were also determined. From the results of these determinations the data presented in Table 21 were collected. In this table are given the maximum, minimum, and average weights per gallon of oysters before and after washing, the computed gain in weight, and the computed gain in volume, the loss in solids, and the computed quantity of water added during washing.

TABLE 21.—*Changes in volume, weight, and solids content of oysters during washing processes*

Experiment	Weight per gallon					
	Before washing			After washing		
	Maximum	Minimum	Average	Maximum	Minimum	Average
	Ounces	*Ounces*	*Ounces*	*Ounces*	*Ounces*	*Ounces*
Tub washing No. 5:						
First soaking period	137	134.79	135.82	136.31	133.33	134.89
Second soaking period	----------	----------	----------	136.4	134.12	135.11
Tub washing No. 6:						
First soaking period	138.44	135.15	136.62	136.23	133.62	134.84
Prolonged soaking period	139	135.42	137.4	136.12	133.5	134.6
Blower washing No. 5	138.41	136.51	137.42	136.89	135.08	135.82
Blower washing No. 6	137.59	135.26	136.28	137.15	134.03	135.35

Experiment	Volume of entire lot				Weight of entire lot			
	Before washing	After washing	Gain		Before washing	After washing	Gain	
	Gallons	*Gallons*	*Gallons*	*Per cent*	*Ounces*	*Ounces*	*Ounces*	*Per cent*
Tub washing No. 5:								
First soaking period	25	27	2	8.0	3,395.5	3,642.11	246.61	7.3
Second soaking period	----------	28.1	1.35	5.05	----------	3,790.53	189.23	5.2
Total soaking period	25	[1] 28.35	3.35	13.4	3,395.5	[2] 3,831.34	435.84	12.8
Tub washing No. 6:								
First soaking period	25	27.6	2.6	10.4	3,415.57	3,721.48	305.91	9.0
Prolonged soaking period	25	29.7	4.7	18.8	3,435.23	3,997.7	562.47	16.4
Blower washing No. 5	20	21	1	5.0	2,748.48	2,850.39	101.91	3.7
Blower washing No. 6	20	20.6	0.6	3.0	2,725.67	2,783.56	57.89	2.1

Experiment	Solids in sample		Solids in entire lot			
	Before washing	After washing	Before washing	After washing	Loss	
	Per cent	*Per cent*	*Ounces*	*Ounces*	*Ounces*	*Per cent*
Tub washing No. 5:						
First soaking period	13.37	10.91	453.98	397.35	56.63	12.5
Second soaking period	----------	9.72	----------	396.02	24.5	6.2
Total soaking period	----------	----------	453.98	[1] 372.41	81.57	18.0
Tub washing No. 6:						
First soaking period	14.43	11.43	492.87	424.37	67.50	13.7
Prolonged soaking period	15.35	10.76	527.31	430.15	97.16	18.4
Blower washing No. 5	----------	----------	[2] 452.67	390.50	62.17	13.7
Blower washing No. 6	----------	----------	347.25	316.49	30.76	8.9

Experiment	Solids per average gallon				Water added during washing	
	Before washing	After washing	Loss			
	Ounces	*Ounces*	*Ounces*	*Per cent*	*Ounces*	*Per cent*
Tub washing No. 5:						
First soaking period	18.16	14.72	3.44	18.9	303.24	8.9
Second soaking period	----------	13.13	1.59	10.8	213.76	5.9
Total soaking period	18.16	13.13	5.03	27.7	517.41	15.2
Tub washing No. 6:						
First soaking period	19.72	15.41	4.31	21.8	373.41	10.9
Prolonged soaking period	21.09	14.48	6.61	31.3	659.63	19.2
Blower washing No. 5	22.63	18.62	4.01	17.7	164.08	5.8
Blower washing No. 6	17.36	15.34	2.02	11.6	88.65	3.2

[1] Corrected to include one-fourth gallon (34.81 ounces) withdrawn for sample.
[2] Calculated from the total solids content found by analysis.

Although in tub-washing experiment 5, 12.5 per cent of the solids was lost during the first washing period, about three-fourths of this consisted of insoluble material, such as shell, sand, and shreds of torn meat. The quantity of solids abstracted by the second washing was smaller than that removed during the first washing, but the actual loss of oyster solids was greater. This is due to the fact that there was very little insoluble material in the 24.5 ounces of solids abtracted by the second washing. In this experiment about 50 per cent of the salt was removed in the first washing and nearly all of it in the second washing. In tub-washing experiment 6 the increases in volume and weight and the loss of solids are somewhat greater than those in experiment 5. This may be explained by the difference in the quality of the oysters. The results of these experiments indicate that an ordinary commercial washing causes gain in volume, addition of water, and removal of solids. Practically all the insoluble solids are removed by such ordinary washing. It is concluded that excessive washing is not needed to remove sand and pieces of shell. Prolonged washing caused a progressive reduction of solids. Very little free liquor developed on the oysters after standing, indicating that much of the added water was retained.

The main differences in procedure between the two washing experiments with a blower (No. 5 and No. 6) and the tub-washing experiments, except that a blower is used, were that only one washing period was used in each experiment and no figures covering the analyses of the wash water were collected. Both blowing experiments were alike except that in the first the time of blowing was five minutes and in the second seven minutes. The oysters used in the second blowing experiment were of poor quality and had recently been frozen.

In both sets of experiments there were gains in volume and weight, addition of water, and loss of solids, the latter being due to a leaching out of solids and not to an apparent decrease from dilution with water. (Tables 22 and 23.) The results emphasize the fact that there is only a small increase in volume when oysters of poor quality are soaked, but a marked loss of valuable oyster solids through leaching.

TABLE 22.—*Quantity of solids present in wash water and derived from oysters in tub-washing experiments*

Experiment	Solids in hydrant water	Soluble solids in wash water	Soluble solids derived from oysters	Soluble oyster solids in wash water	Solids lost during washing	Insoluble solids removed by washing [1]
	Grams per liter	*Grams per liter*	*Grams per liter*	*Ounces*	*Ounces*	*Ounces*
No. 5:						
First soaking period	0.53	3.08	2.55	16.32	56.63	40.31
Second soaking period	.53	2.76	2.23	21.92	24.54	2.62
No. 6:						
First soaking period	.09	5.19	5.1	32.28	67.50	35.22
Prolonged soaking period	.09	5.01	4.92	46.2	97.16	50.96

[1] Insoluble solids consisted of shell, sand, shredded meat, etc.

TABLE 23.—*Effect of washing on composition of oysters*

Method and material	Free liquor	Meat	Liquor	Solids	Salt	Meat		Liquor		Entire sample		Loss in solids
						Solids	Salt	Solids	Salt	Solids	Salt	
	Per cent	*Per cent*	*Per cent*	*Grams per liter*	*Grams per liter*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
Tub washing No. 6:												
Oysters before washing	(1)	(2)	(2)			13.37	0.11			13.37	0.11	0
Oysters after first washing	(2)	91.4	8.6			11.61	.05	3.48	0.15	10.91	.06	18.4
Oysters after second washing	(2)	91.8	8.2			10.25	(4)	3.74	.08	9.72	(4)	27.3
Two 1-gallon cans of soaked oysters	(3)	94.3 93.6	5.7 6.4									
Hydrant water				0.55	(4)							
First wash water				3.03	0.48							
Second wash water				2.76	.22							
Tub washing No. 6:												
Oysters before 2-hour washing	(2)	90.9	9.1			15.40	.18	4.69	.61	14.43	.22	0
Oysters after 2-hour washing	(2)	90.8	9.2			12.21	.04	3.71	.21	11.43	.06	20.8
Oysters before 4-hour washing	(2)	91.8	8.2			16.32	.27	4.47	.81	15.33	.31	0
Oysters after 4-hour washing	(2)	89.8	10.2			11.02	(4)	3.18	.16	10.76	.02	29.9
Hydrant water				.09	(4)							
2-hour wash water				5.19	1.15							
4-hour wash water				5.01	1.25							
Blower washing No. 5:												
Oysters before blowing	2.2									16.47	.08	0
Oysters after blowing	4.6									13.71	.03	16.8
Blower washing No. 6:												
Oysters before blowing	6.8									12.74	.18	0
Oysters after blowing	4.9									11.37	.06	10.8

[1] Almost none visible. [2] Not separated. [3] Small quantity. [4] Trace. [5] None.

In all of the washing experiments so far discussed the computation of water added has been made on the basis of gain in volume and in weight rather than on the basis of the difference between the solids in the original material and those in the washed material. In blower experiment 5, analyses of the oysters before and after washing showed a loss in solids of 16.8 per cent. Weighings before and after blowing, however, showed that these oysters did not take up 16.8 per cent of water but that the quantity of water actually incorporated was about 6 per cent. The fallacy of computing water added by solids lost is apparent, as it is based on the supposition that the apparent reduction in solids is due entirely to dilution with water and does not take into account the actual loss of soluble solids removed in the wash water.

WHAT CONSTITUTES GOOD WASHING

During any effective washing process there is some gain in volume and some loss in solids and salt of shucked oysters. These changes depend upon so many factors that it is difficult to conclude, in one general statement, just what constitutes a proper washing without adulteration with water. The following conclusions, however, can be applied within certain limits:

1. In oysters subjected to a reasonable washing with fresh water, gains in volume from 3 to 10 per cent or more may take place, depending upon the original condition of the oysters, whether fat or

lean, upon the apparatus used, and upon the duration of the washing period. At the same time there may be losses in solids varying from 8 or 9 to 13 or 14 per cent or more. Usually more than half of the salt in the oysters is removed. The use of a blower causes less gain in volume and a smaller loss in solids than the use of tubs, provided the blowing is not continued too long and the oysters are not allowed to remain in the wash water after the blowing period is ended. Furthermore, the use of a weak brine (about 0.5 to 1.5 per cent salt solution) results in reducing the volume and causing an apparent increase in solids content.

2. When the oysters are subjected to a prolonged washing, which actually constitutes a soaking process, there is a further gain in volume and loss in solids and salt. During such a soaking process the loss in solids is mostly a removal of soluble solids, as the shell pieces, sand, etc., are removed early in the washing process. In other words, a prolonged washing is not necessary to remove objectionable insoluble matter. Once such material is removed the additional washing continues to remove solids from the oysters.

3. Solids are removed from the oysters by the wash water. (Table 23.)

4. The greater part of the water absorbed by oysters is retained during shipping and subsequent standing.

In addition to increasing the volume of the oysters, adding water, and removing valuable solids, excessive washing takes away the sea tang and fine flavor which has long placed oysters at the top of the list of popular sea foods. Continued contact with fresh water destroys their more desirable physical characteristics. The rich, creamy yellow color and firm texture disappear and the oysters become chalky white, bloated, and puffed, with soft, spongy tissues. On cooking, such oysters rapidly diminish to less than their original size and the meat becomes tough and almost devoid of taste. Their nutritive value is necessarily lowered by soaking in fresh water, which removes large quantities of the soluble nitrogenous compounds and carbohydrates. It seems probable that their digestibility is also lowered, as the remaining solids are the tougher muscular and fibrous tissues, which probably are less easily digested than the more soluble ingredients removed by excessive washing.

Under the terms of the Federal food and drugs act (*91*), it was the duty of the Bureau of Chemistry to prevent the sale of adulterated foods and to protect the consuming public from fraud. Oysters lend themselves readily to adulteration with water. It is very difficult to draw a line between proper washing, which will produce a clean, marketable foodstuff, and soaking, which brings about adulteration with water. At the same time washing is a necessary operation, which can not be done without some loss of food constituents and addition of water.

From the bacteriological standpoint, the best cleansing is performed by the use of a blower. From the chemical standpoint, the use of a blower for about three minutes, a weak brine (about 0.5 per cent) being used as a washing medium, produces the least change in volume and solids content. The most efficient and safest method of washing oysters is by using a blower with salt water.

SHIPPING OYSTERS

CAUSE OF DECOMPOSITION

Products having a high water content invariably spoil more rapidly than drier materials. As the development of bacteria and other microorganisms responsible for the decomposition of foods is favored by the presence of moisture, any food containing a high percentage of water is extremely perishable. Therefore, in handling and shipping oysters precautions to prevent spoilage of the product between the time of shipment and the time of consumption are necessary.

In this connection it is not illogical to compare oysters with milk. Both decompose rapidly if not handled properly. Frequently each is consumed in the raw state and each is capable of supporting the growth of disease-producing bacteria unless precautions are taken to prevent the access of such organisms to the food or to eliminate them. The hygienic principles of handling ordinarily applied to milk apply to shucked oysters.

Oysters in the shell will remain in good condition as long as they live. By means of its strong adductor muscle an oyster out of water keeps its shell closed tight most of the time, although occasionally oysters in a sack or barrel open their shells slightly during shipment or storage. If still alive such oysters close their shells quickly when disturbed. When the oyster dies the adductor muscle is relaxed and the shell remains open. Dead oysters decompose rapidly, one decomposing oyster serving as a source of contamination to the entire lot in the shipping container. Kept in a cool place shell oysters will remain alive and in edible condition for comparatively long periods.

The real bacteriological problem in shipping oysters is met in the handling and storage of shucked oysters subsequent to their treatment in the shucking house.

In products having a high carbohydrate content the predominating type of spoilage is fermentation. In protein products, such as meat and fish, it is putrefaction. Oysters contain both protein and carbohydrate (glycogen), so that during decomposition fermentation as well as putrefaction takes place. After the death of the oyster, which occurs soon after shucking, the glycogen present is hydrolyzed to produce reducing sugars. These sugars are readily fermentable by bacteria of many species. The fermentation is productive of acid, mainly lactic acid, and "sour" oysters. Although the production of gas, acid, and a characteristic odor in decomposed oysters is responsible for their designation as sour oysters, putrefaction is also taking place.

An investigation by Hunter and Linden (*48*) showed that no relation exists between the total number of aerobic bacteria present and the condition of the oysters. As many as 30,000,000 aerobic bacteria per cubic centimeter of oyster liquor were found in oysters that were in good condition, as far as decomposition was concerned. Only 12,000 aerobic bacteria per cubic centimeter of liquor were found in others considered, because of odor and appearance, to be in an incipient state of decomposition. The factors involved influence the bacterial count tremendously. If oysters taken during the hibernating season are studied, the number of aerobic bacteria present is much smaller than when nonhibernating oysters are studied. The

introduction, during shucking, of bacteria that are not removed by washing provides a high count of microorganisms, even when the oysters are fresh and in good condition. It is, therefore, futile to attempt to grade oysters as to decomposition by the use of bacterial counts. It was apparent from this study that the spoilage of oysters depends upon the presence and development of bacteria of certain types or groups rather than upon the total number of organisms present. It was evident that the total number of aerobic bacteria present was not as significant in judging the quality of shucked oysters as might be the numbers of bacteria of certain groups or species which cause fermentation or putrefaction.

Further experiments to determine the types or groups of bacteria responsible for decomposition of shucked oysters showed that certain bacteria isolated from decomposing shucked oysters, when introduced in pure culture into an oyster medium, produced foul, putrefactive odors. These organisms were identified as members of the genera Serratia[1] (water and soil bacteria producing red pigment), Pseudomonas (soil and water bacteria producing a blue-green pigment), Proteus, Clostridium (spore-forming obligate anaerobes), and Bacillus (aerobic spore-forming bacteria). Certain other microorganisms produced acidity or sour odor, or both, in pure cultures grown in oyster medium. The microorganisms were members of the lactose-fermenting group of bacteria, such as *Aerobacter aerogenes*, *A. cloacae* and *Escherischia coli*, lactobacilli, streptococci, and yeasts. A great many other water and soil bacteria, which apparently had no effect upon oysters, were isolated from the decomposing material. The majority belonged to the genera Achromobacter, Eberthella, and Flavobacterium.

A study indicated that the decomposition of shucked oysters in the beginning is due to the activities of some members of the Serratia, Pseudomonas, Proteus, Clostridium, Bacillus, Aerobacter, and Escherischia groups of bacteria. Later in the course of the spoilage streptococci, lactobacilli, and yeasts find more suitable conditions for development, until in the very late stages of decomposition the high dilution plate cultures made from the oysters, which became very sour and putrid, contained almost exclusively colonies of these three groups of organisms.

The information now at hand, which may be changed by further research, indicates that both fermentation and putrefaction take place during the decomposition of oysters and that the spoilage is due to the action of ordinary water and soil bacteria, with such intestinal bacteria as may be present from pollution with sewage.

RATE OF DECOMPOSITION

The rapidity with which shucked oysters decompose depends upon the conditions under which they are handled. The fact that shucked oysters are a perishable product means that spoilage advances very quickly unless precautions are taken to prevent it. The results of laboratory experiments to determine the effect of washing in fresh water and brine and of the use of ice upon the keeping quality of shucked oysters are given in Table 24.

[1] The nomenclature for bacteria as presented in Bergey's Manual of Determinative Bacteriology by a committee of the Society of American Bacteriologists (80) is used.

TABLE 24.—*Relation between treatment of oysters and time and progress of spoilage*

Jar No.	Treatment of oysters	Day of storage on which oysters became stale	Day of storage on which oysters became sour
EXPERIMENT 1 (OYSTERS CONSTANTLY IN ICE; TEMPERATURES 33 TO 55° F.)			
1	Shells unwashed; oysters unwashed	3	10
2	Shells unwashed; oysters washed in fresh water	4	12
3	Shells unwashed; oysters washed in brine	4	14
4	Shells washed; oysters unwashed	3	14
5	Shells washed; oysters washed in fresh water	4	14
6	Shells washed; oysters washed in brine	4	14
EXPERIMENT 2 (NO ICE USED; TEMPERATURES 57 TO 74° F.)			
7	Shells unwashed; oysters unwashed	1	3
8	Shells unwashed; oysters washed in fresh water	2	4
9	Shells unwashed; oysters washed in brine	2	4
10	Shells washed; oysters unwashed	2	4
11	Shells washed; oysters washed in fresh water	2	3
12	Shells washed; oysters washed in brine	2	3
EXPERIMENT 3 (ICED ON ALTERNATE DAYS; TEMPERATURES 46 TO 76° F.)			
13	Shells unwashed; oysters unwashed	2	8
14	Shells unwashed; oysters washed in fresh water	2	7
15	Shells unwashed; oysters washed in brine	2	7
16	Shells washed; oysters unwashed	2	8
17	Shells washed; oysters washed in fresh water	2	7
18	Shells washed; oysters washed in brine	2	7
EXPERIMENT 4 (NO ICE USED; TEMPERATURES 49 TO 65° F.)			
19	Shells unwashed; oysters unwashed	2	4
20	Shells unwashed; oysters washed in fresh water	2	3
21	Shells unwashed; oysters washed in brine	2	4

Apparently the washing, before and after shucking, had little effect upon the keeping quality. The oysters became stale or sour on about the same day, regardless of the method of washing. Where ice was used constantly the oysters developed a slightly abnormal odor on the third and fourth days, but remained in this condition until the tenth, twelfth, and fourteenth days. These oysters did not, in the late stages of decomposition, develop the characteristic sour odor and gassy appearance of badly decomposed oysters, but developed a strong, rank, disagreeable odor and a milky appearance.

In cases where no ice was used the oysters changed very rapidly from normal to a condition described as stale and within three or four days after shucking to a sour-smelling, nauseating, gassy, and milky appearing condition, unmistakably characteristic of spoiled oysters.

The use of ice on alternate days simply retarded temporarily the growth of the bacteria causing spoilage, but the progress of the decomposition on the days when no ice was used was so rapid that the spoilage proceeded gradually, regardless of the temperature. A condition of sourness was reached on the seventh or eighth day. It is evident that rapid spoilage takes place unless oysters are held at the

proper refrigerating temperatures. If shucked oysters are handled properly during shipment and storage, they can be shipped from the Atlantic coast to points in the Middle West.

DETECTION OF SPOILAGE

Probably the best method of detecting spoilage in shucked oysters is by the use of the senses of smell and sight; that is, the so-called organoleptic test. Decomposed oysters have a characteristic odor and appearance that is unmistakable to any one familiar with the fresh product. Certainly no other test is needed for the detection of spoilage in oysters that have reached an advanced stage of decomposition. It is not unusual, however, to find shucked oysters that have passed from the perfectly fresh stage to the incipient stages of decomposition. In judging the quality of such oysters, which, for the lack of a better word, have been designated as "stale," there is opportunity for personal opinion, likes, and dislikes to play a part. In view of this, Hunter and Linden (*48*) attempted to correlate some definite laboratory test with the physical condition of the oysters. An attempt to establish a relationship between the total counts of bacteria and the condition of the oysters failed (p. 57). A study of the relationship between the hydrogen-ion concentration of oyster liquor and the stage of decomposition was more productive of results. The hydrogen-ion concentration of the oyster liquor was determined, as accurately as the method would permit, by testing, in a porcelain plate, a drop of the liquor with certain hydrogen-ion indicators described by Clark and Lubs (*28*). The results of these tests are given in Table 25.

TABLE 25.—*Limiting pH values for good, stale, and sour oysters*

EXPERIMENT 1

Jar No.	Treatment	pH values			
		Good oysters	Stale oysters	Slightly sour oysters	Sour oysters
1	Shells unwashed; oysters unwashed	6.8–6.05	6.05–5.3	5.3–5.0	5.0 –4.0
2	Shells unwashed; oysters washed in fresh water	6.9–6.15	6.15–5.5		5.5 –5.0
3	Shells unwashed; oysters washed in brine	6.2–6.0	6.0 –5.25		5.25–5.0
4	Shells washed; oysters unwashed	6.5–6.15	6.15–5.4	5.4–5.3	5.3 –5.2
5	Shells washed; oysters washed in fresh water	6.6–6.05	6.05–5.2	5.2–5.1	5.1
6	Shells washed; oysters washed in brine	6.3–6.0	6.0 –5.2	5.2–5.0	5.0

EXPERIMENT 2

7	Shells unwashed; oysters unwashed	7.0–6.6	6.6 –5.0		5.0 –4.7
8	Shells unwashed; oysters washed in fresh water	6.8–6.15	6.15–4.75		4.75–4.7
9	Shells unwashed; oysters washed in brine	6.7–5.8	5.8 –4.8		4.8 –4.7
10	Shells washed; oysters unwashed	7.0–6.25	6.25–4.9		4.9 –4.7
11	Shells washed; oysters washed in fresh water	6.8–5.9	5.9 –4.75		4.75–4.7
12	Shells washed; oysters washed in brine	6.6–6.1	6.1 –4.9		4.9 –4.7

TABLE 25.—*Limiting pH values for good, stale, and sour oysters*—Continued

EXPERIMENT 3

Jar No.	Treatment	pH values			
		Good oysters	Stale oysters	Slightly sour oysters	Sour oysters
13	Shells unwashed; oysters unwashed	7.0–6.5	6.5–5.2		5.2–4.7
14	Shells unwashed; oysters washed in fresh water	6.8–6.5	6.5–5.4		5.4–4.7
15	Shells unwashed; oysters washed in brine	6.6–6.3	6.3–5.2		5.2–4.7
16	Shells washed; oysters unwashed	6.8–6.4	6.4–5.3		5.3–4.7
17	Shells washed; oysters washed in fresh water	6.6–6.3	6.3–5.3		5.3–4.7
18	Shells washed; oysters washed in brine	6.5–6.25	6.25–5.3		5.3–4.7

EXPERIMENT 4

19	Shells unwashed; oysters unwashed	6.9–5.8	5.8–5.1		5.1–4.7
20	Shells unwashed; oysters washed in fresh water	6.8–5.6	5.6–5.0		5.0–4.7
21	Shells unwashed; oysters washed in brine	6.8–5.6	5.6–4.9		4.9–4.6

The limiting hydrogen-ion concentration values, which were fairly definite, may be of some assistance in determining the quality of shucked oysters. A hydrogen-ion concentration value between 5.6 and 6.1 apparently represents a zone wherein oysters are passing from good to stale. Oysters passing from stale to sour or putrid had hydrogen-ion concentration values between 4.9 and 5.3. Oysters having a hydrogen-ion concentration of less than 5.0 may be considered usually to be in an advanced stage of decomposition. The limited number of determinations and the restricted area from which the oysters used were collected do not justify the unqualified statement that these hydrogen-ion concentration values may be applied to shucked oysters from all localities handled under all conditions. The results obtained, however, were consistent enough to indicate that the hydrogen-ion concentrations given are significant and may be of value in examining shucked oysters of questionable quality.

PREVENTION OF SPOILAGE

In order to prevent spoilage during shipment shucked oysters must be kept at temperatures low enough to prevent, or at least to retard, the development of microorganisms (*15*, *71*). Shucked oysters shipped and stored at temperatures below 50° F., preferably below 45°, should reach the consumer in good condition. Washing has a decided effect upon the quality of shucked oysters, especially on the number of organisms present (p. 47), but it is impossible to free oysters entirely of bacteria by washing (p. 47). The two main factors in the prevention of spoilage are the avoidance of contamination and the liberal use of ice.

Modern methods of shipping shucked oysters in nonreturnable containers have done away with the objectionable practice of placing ice in contact with the oysters. Oysters are now packed "dry" in 1, 2, or 5 gallon tin cans fitted with friction tops and in quart and pint containers. These cans, which are cylindrical, are placed in rectangular or square wooden boxes or in barrels. Below and above the can, and in the corners around it, is placed plenty of crushed ice, which does not come in contact with the oysters but keeps them

cooled to the desired temperature if they are re-iced, as they should be, during transit. Unless shucked oysters are to be kept in a refrigerator or cold-storage plant in the retail store, the same principle of refrigeration should be applied there as during shipment. Ice should not be added directly to the oysters, but they should be kept in suitable metal containers surrounded by ice. Carelessness on the part of the retailer may nullify all the efforts which have been taken in preparing the product.

SIGNIFICANCE OF "FREE LIQUOR"

In commercial practice there is always the possibility that draining and skimming may not be properly carried out and that quantities of wash water may be carried into the shipping container to adulterate the oysters during shipment and storage. Many experiments were conducted in the Bureau of Chemistry to determine the effect of adding known quantities of water to shucked stock in the shipping container. For example, four experiments were undertaken to ascertain whether or not properly skimmed oysters developed free liquor during shipment or storage and to determine how far oysters if adulterated with fresh water, either through deliberate intent or as a result of improper draining, would absorb this excess of liquid during storage or shipment.

In each experiment properly skimmed, "dry" oysters were packed in 1-gallon friction-top cans. Other cans were filled with oysters from the same batch, known quantities of water being added. In each experiment four sets of two cans each were prepared. (Table 26.) In the first and fourth experiments hydrant water was used as an adulterant. In the second and third experiments wash water from a blower was used, as it was assumed that improperly drained oysters would be adulterated with such a solution of oyster solids. The cans in the first experiment were shipped direct from Norfolk, Va., to Baltimore, Md. Those in the other three experiments were shipped from Crisfield, Md., and from Norfolk, Va., to Baltimore, Md., via Buffalo, N. Y., where they were re-iced.

In examining the oysters after shipment a record was taken of their appearance and determinations were made of the quantity of free liquor and the percentage of solids and salt in the entire sample. The data collected are presented in Table 26.

TABLE 26.—*Appearance of and quantity of free liquor, solids, and salt in washed and drained shucked oysters to which water was added*

"DRY" OYSTERS (BACK RIVER AND MOBJACK BAY STANDARDS)

Experiment No.	Method of treating	Condition after shipment				
		Appearance	Free liquor	Solids[1]	Salt[1]	Decrease in solids
			Per cent	*Per cent*	*Per cent*	*Per cent*
1	No water added	Solid pack; little free liquor on the surface.	0.09 1.2	14.52	0.17	0.0
	Plus 5 per cent hydrant water	do	2.4 2.0	13.76	.16	5.2
	Plus 10 per cent hydrant water	Solid pack; some free mucilaginous liquor on surface	4.6 2.2	13.20	.15	9.1
	Plus 15 per cent hydrant water	do	5.1 3.5	12.48	.14	13.9

[1] Determinations made on entire sample.

TABLE 26.—*Appearance of and quantity of free liquor, solids, and salt in washed and drained shucked oysters to which water was added*—Continued

"DRY" OYSTERS (GREAT WICOMICO STANDARDS)

Experiment No.	Method of treating	Condition after shipment				
		Appearance	Free liquor	Solids [1]	Salt [1]	Decrease in solids
			Per cent	*Per cent*	*Per cent*	*Per cent*
[2]	No water added	Some free liquor on surface	4.6 7.4	13.70	0.03	0.0
	Plus 10 per cent water taken from blower after washing.	Free liquor, about 1 inch deep, on surface.	13.1 18.7	12.0	.03	7.4
	Plus 15 per cent water taken from blower after washing.	Large quantity of free liquor; sloppy pack.	18.5	11.39	.03	16.8
	Plus 20 per cent water taken from blower after washing.	About 2 inches of free liquor on surface; sloppy pack.	24.5 23.5	10.78	.03	21.3

"DRY" OYSTERS (YORK RIVER STANDARDS)

3	No water added	Solid pack; very little free liquor visible.	5.7 6.9	11.1	0.04	0.0
	Plus 10 per cent water taken from blower after washing.	Some free liquor on surface	11.8 11.0	10.2	.03	8.1
	Plus 15 per cent water taken from blower after washing.	Much free liquor; somewhat sloppy.	15.7 17.2	9.5	.03	14.4
	Plus 20 per cent water taken from blower after washing.	Large quantity of free liquor; sloppy pack.	20.5 20.4	9.0	.03	18.9

"DRY" OYSTERS (JAMES RIVER SELECTS)

4	No water added	Free liquor, about ¼ inch deep, on surface.	6.6	12.36	0.09	0.0
	Plus 10 per cent hydrant water.	Free liquor, about 1 inch deep, on surface.	12.4	11.91	.12	3.64
	Plus 15 per cent hydrant water.	Free liquor, about 1½ inches deep, on surface.	20.7	10.9	.12	11.9
	Plus 20 per cent hydrant water.	Free liquor, about 2 inches deep, on surface.	24.0	10.23	.13	17.2

[1] Determinations made on entire sample.
[2] These oysters were frozen between packing and examination.

In experiments 2 and 3 the added wash water was not absorbed as in experiment 1, where the maximum quantity of free liquor was 5.1 per cent in the samples adulterated with 15 per cent added water. This may be explained by the use as an adulterant, in experiments 2 and 3, of a liquid which already contained oyster solids and was not easily absorbed, and by the fact that the oysters in these experiments were of low solids content.

Several conclusions may be drawn from these experiments. If plump, fat oysters of fairly high solids content are used, a reasonable quantity of water added as an adulterant will be absorbed and retained, not to be given up as free liquor upon storage. The added water will dissolve oyster solids and will become, in fact, a thick, viscous solution of soluble oyster material. The detection of such adulteration by chemical analysis is difficult. If lean oysters of fairly low solids content are used, the added water is not absorbed and can readily be detected as free liquor after storage. In such cases the quantity of free liquor is proportionate to and may equal the quantity of water added. Even oysters to which no water has been intentionally added will develop some free liquor upon standing, but this is rarely in excess of 5 per cent by weight, a fair average for the quantity of free liquor on properly washed and packed

oysters. When only small quantities of water are added a higher proportion is absorbed than when large quantities are added. Data at hand indicate that oysters showing free liquor in excess of 10 per cent by volume are adulterated with water. As oysters do not normally develop much more than 5 per cent of free liquor, it is safe to state that 10 per cent indicates adulteration. As the solids, salt, and moisture contents vary greatly with the locality and with the season of the year, making it difficult always to detect adulteration with water by moisture and salt determinations, it is probable that the examination of shucked oysters for free liquor is a more reliable means of detecting adulteration. At least, such determinations are confirmatory of results obtained by examining them for total solids and salt.

GREEN OYSTERS

Probably no phase of the biology of the oyster has been subject to more controversy than has the subject of green oysters. Some of the early investigators stated that the green coloration in oysters was due to the presence of copper. Others contradicted this statement. This diversity of opinion is explained, to a great extent, by the fact that the oysters used for study were from different sources and that the green color was not the same in all the oysters used. When the existence of more than one type of green oysters was finally established there appeared to be more agreement among biologists regarding the cause of each type.

It is now known that there are two distinct types of green oysters. In one the color is distributed in streaks and patches over the liver or the visceral part of the body. Sometimes the entire body has a greenish tinge. This light bluish green, suggestive of the color of certain copper salts, led to investigations to prove that it was due to the presence of copper in the body of the oyster.

In the other type of green oyster the color is restricted to the gills and mantle. The coloration in such oysters is usually dark olive green, somewhat suggestive of chlorophyll. Oysters exhibiting this condition are referred to as green gilled, green bearded, or green finned.

COLORED BY COPPER

Prior to 1894, many of the investigations reported in the literature appear to have been conducted as attacks upon the popular theory that the greening of oysters was due to copper. In 1894, Bulstrode (*21*) reported that oysters from Falmouth and Truro, England, which showed the bluish-green coloration in patches, were entirely different from those from Marennes, France, in which the green color was restricted to the gills or mantle. Samples of the blue-green oysters were collected by Bulstrode, who sent them to Thorpe for analysis. Thorpe (*84*) stated that "there is no question that the greenness of certain oysters, especially of those found in Falmouth and Truro waters, is due to copper." Herdman and Boyce (*42*) found copper in quantities up to four times the normal in green American oysters. The copper reaction coincided histologically with the presence of green granular leucocytes. Consequently, the copper was regarded as the cause of the green color. The investigators also found that the copper content of normal "white" European oysters varied

from 0.25 to 0.66 milligram per oyster, with an average of about 0.4 milligram. Oysters exhibiting green patches on the body contained 3.52 milligrams of copper, about nine times that found in the normal oyster. Green American oysters contained 2.63 milligrams of copper per oyster; normal oysters from the same source contained only 0.7 milligram of copper. By microchemical methods Herdman and Boyce demonstrated the presence of excessive quantities of copper in the green tissues of American oysters. Experiments in which certain metallic salts were added to the water in which oysters were floated were not successful in producing the green color, although there were some indications of green color in the floated oysters. As a result of their work, Herdman and Boyce were positive that the bluish-green color appearing in spots on the body of the American and English oyster was due to copper.

Pease (*69*), Nelson (*65*), and some others were equally positive that this green color was not due to copper. Pease reported that all oysters contain some copper and that of 60 samples examined in his laboratory the copper content varied from 2 to 782 milligrams per 1,000 grams. The smallest quantity of copper in green oysters was 107 milligrams per 1,000 grams of oyster and the greatest in normal oysters was 295 milligrams per 1,000 grams. In other words, normal oysters sometimes contain more copper than do green oysters. Pease stated that high copper content and greening always appear together in oysters, the average copper content for all green oysters being 391 milligrams per 1,000 grams and the average for all normal oysters only 49 milligrams per 1,000 grams. Nelson believed that the green color was not due to copper, even though he found in green oysters four or five times as much copper as was found in normal oysters.

In 1917, Stewart (unpublished report) stated that the bluish-green color which permeates the entire body of the oyster is undoubtedly due to the presence of copper in excess of the normal amount.

The preponderance of the evidence reported in the literature favors the view that the bluish-green color in patches on the bodies of oysters is due to the presence of an excessively large quantity of copper. The source of the copper in the sea water can not be satisfactorily explained. It has been suggested that it is derived from trade wastes, from the copper bottoms of vessels, from drainage through copper-bearing soil, and from other sources. Regardless of the source, however, there is no doubt that comparatively large quantities occur in green oysters. This green color is not easily removed from the body of the shellfish. Green oysters transplanted to other waters than those in which they were grown remain green for long periods of time.

The question of injury to health from the consumption of oysters containing copper is also an open one. Many investigators have claimed that the quantity of copper present is too minute to be injurious unless a great many more oysters are eaten than are usually consumed by one person. The principal objections to such oysters have been their appearance and peculiar taste. Stewart (unpublished report) stated that he had eaten oysters containing as much as 1,000 parts per million of copper with no apparent discomfort. However,

it can not be recommended that oysters containing much copper be eaten. Until more information is obtained regarding possible danger to health from copper-green oysters, it is probably well to view such shellfish with suspicion.

GREEN GILLED

Oysters exhibiting a dark-green coloration in the gills and mantle have been known and relished for a long time in France. Such oysters, called "huitres de Marennes," were produced artificially in tanks or "claires," as they brought a higher price in the French market than did the normal white oysters. In comparatively recent years green-gilled oysters have been recognized in the United States, but not with the favor shown them in Europe. In the United States green-gilled oysters are usually viewed with suspicion by the consumer and hence are almost unmarketable.

As early as 1820 Gaillon (*38*) published a report of his studies on green-gilled oysters. Later many biologists, including Puységur (*72*), Ryder (*78*), Lankester (*59*), Bulstrode (*21*), and Herdman and Boyce (*42*) confirmed Gaillon's conclusion that the green coloration in the gills of the oyster is due to the presence of the diatom *Navicula ostrearia*.

In the fall and early winter of 1915, the appearance of green-gilled oysters in Lynnhaven Bay, Va., led the United States Bureau of Fisheries to conduct investigations to learn whether or not these oysters were identical with those of Marennes. Mitchell and Barney (*63*) concluded that the Chesapeake green-gilled oyster was the same as the Marennes oyster.

Apparently investigators agree that the color is due to the ingestion of diatoms by oysters, not to the presence of iron or copper. They agree also that such greening does not make the oysters injurious to health and that, if the prejudice of the public for such an abnormal-appearing product can be overcome, there is no reason why green-gilled oysters should not find as ready a market as white oysters. The sanitary quality of the oysters can not be correlated with the green color of the gills, as this color may appear in fresh, wholesome shellfish from unpolluted sources. In the United States it is simply a case of nature producing what is sought by artificial means in France.

As it is well established that the color in green-gilled oysters is due to microscopic plants, which are not injurious when eaten, and as the presence of this color in no way indicates that the oysters are either decomposed or polluted, there can be no objection to the sale of green-gilled oysters because they are green. The standards for decomposition and pollution that are applied to all other oysters should be applied to green oysters, no matter whether their color is due to copper or to the presence of the diatom *Navicula fusiformis* var. *ostrearia*.

PINK OYSTERS

The production of a pink color in shucked oysters during transit or storage, even at low temperature, is frequently the cause of serious financial loss to shippers and dealers. In 1914, Bates and Round suggested that this pink color was produced by a yeastlike fungus.

A detailed report of the cause of the pink color, with suggestions for its control, was later presented by Hunter (*47*).

Upon reaching their destination oysters shipped from points on the Atlantic coast to the Middle West often show a coral pink or reddish color in the liquor or on the meat. Sometimes this color is not evident to the jobber or distributor who receives the oysters about two days after shucking and who repacks and reships them to retailers in his territory. Neither does the color appear immediately after shucking and before shipping. Investigation demonstrated that the color was due to the development of a pink yeast, which grows readily and produces pigment at low temperatures. The color produced in the oysters has been variously reported as a bright red or a faint pink. The prevailing shade seems to be a coral pink.

After the cause of the color had been determined, studies were conducted to ascertain the source of the contamination and to find some method of control. The pink yeast was found on the bins, benches, tubs, wheelbarrows, and other utensils in and about the oyster-shucking house. It was particularly abundant in the accumulated dust and dirt within the bins. The yeast could be recovered also from oysters, especially after they had been in contact with infected objects. Although the yeast was present in oysters taken directly from the growing areas, the number of such organisms was very small. The results indicated that the chance of contamination was far greater within the shucking house than on the beds. Repeated examinations of samples of surface and bottom water from near the oyster-growing areas rarely revealed the presence of the pink yeast. Samples of water collected while shells from the shucking house were being spread on the beds to serve as cultch for the collection of oyster spat contained the pink yeast. It was indicated that the yeast collected on the shells during handling in the shucking house were being returned to the waters from which oysters were being taken. It was impossible to recover the pink yeast from samples of mud and sand collected from the oyster beds.

The yeast isolated from the pink oysters did not ferment any of the carbohydrates commonly used in the laboratory, nor did it seem to have any effect upon the oysters other than the production of the pink color. The yeast was very resistant to drying. The pink yeast is easily killed by formaldehyde in a dilution of 1 part of formaldehyde gas to 2,500 parts of water. Adding 1 part of commercial formalin, which is a 40 per cent solution of formaldehyde, to 1,000 parts of water gives a 1–2,500 dilution of formaldehyde. As a method of control it is recommended that all bins, benches, tubs, other utensils, and woodwork in the oyster house be scrubbed thoroughly with water and then washed with the formaldehyde solution. The washing should be done in the fall, before any oysters are brought in. The bins, benches, tubs, etc., should also be washed several times during the winter whenever convenient.

When oysters are taken from the water they immediately come in contact with the deck of the boat, which is probably infected with the pink yeast. Oysters are usually placed in tubs, wheelbarrows, or bins that may also be contaminated. During shucking, the oysters become infected with the yeasts from the shells. These yeasts are not easily

removed by the washing process, and are carried, with the oysters, into the shipping container, where they develop and produce the pink color, even though refrigerated.

Pink yeasts are distributed rather widely in nature and are to be expected in places like oyster bins where dust and dirt may remain undisturbed during the summer. Thorough cleansing of the woodwork with a liberal use of formaldehyde solution, however, should remove the cause of the trouble.

Cultures of the yeasts isolated from pink oysters were found to be nonpathogenic to laboratory animals. There is no reason to believe that these yeasts would render shucked oysters injurious. The quality of shucked oysters, with respect to decomposition or possible danger to health, must be judged by criteria other than the presence of a red or pink color. In other words, this color is not a criterion of decomposition. Oysters exhibiting it may or may not be of proper sanitary quality. As the pink color does not appear until several days after shucking, however, it must be assumed that other microorganisms have also developed in the oysters and that by the time the oysters have become pink they are no longer perfectly fresh. The appearance of the pink color indicates that several days have elapsed since shucking and that the oysters are approaching a condition in which they may be considered somewhat stale. Although pink oysters are harmless when eaten and may be edible, they should be examined carefully for evidence of decomposition before being sold or eaten.

Several investigators recently have suggested that there may be another type of pink oysters which owe their color to some agent other than the pink yeast. The development of a pink color in shucked oysters stored at 0° C. and the inability of certain investigators to reproduce the color by inoculation with pink yeasts isolated from the product under examination led to the suggestion that some purely chemical reaction may take place in oysters as a result of which a pink color is produced. At the present time there is no proof of such a phenomenon, but it is not improbable that such may be the case. If future investigations show that a pink color can be produced in oysters without the action of microorganisms, new methods of control and prevention must be developed. In establishments where the control measures here recommended have been applied, however, trouble from pink oysters has ceased.

OLYMPIA OYSTERS

Only one outbreak of typhoid fever has been traced to the consumption of the small native oysters produced on the Pacific coast (*14*). Following a Thanksgiving Day dinner where oyster cocktails were served in a restaurant at San Diego, Calif., many cases of typhoid fever were reported. Investigation indicated that polluted oysters from some small beds in San Diego Bay were used at the dinner. It was stated that "the acute symptoms in the outbreak under consideration were due to oysters having been improperly stored while infected with sewage organisms, and that spoilage permitted of a rapid increase in certain of the bacteria contained." No

case of illness has yet been reported from the consumption of the Olympia oysters produced in the Pacific Northwest.

No special bacteriological method for the examination of these small oysters has been developed. The present standard methods are hardly applicable, as the Olympia oysters are too small to produce enough liquor for the test when only 5 to 12 oysters are used. If a sufficiently large number of such oysters are used in order to obtain the shell liquor necessary for the inoculations there is no reason why the standard procedure should not be followed from that point and the same standards of sanitary quality applied.

Sewage pollution of the oyster beds in the Pacific Northwest has not been a serious factor. The bays in which these oysters are produced are usually fairly remote from centers of population and no large sewers empty into the near-by waters. The Japanese workmen employed to care for oysters and to harvest them usually live in houseboats moored on or near the oyster beds. At one time it was found that waste from these dwellings was polluting the oysters and steps were taken to provide suitable means of waste disposal. Occasional reports received indicate various sources of pollution of the oyster beds, but the volume of sewage in no case has been great and remedial measures have been taken. Very little work, if any, has been done upon the bacterial flora of the Olympia oyster. The statements regarding sanitary surveys and the proper conditions for the growing of oysters (pp. 23–26) are just as applicable to Olympia oysters as to the eastern oysters.

Studies on the hibernation and self-purification of the Olympia oyster have not been reported and no information is available on these questions. There is no reason why Olympia oysters should not function to rid themselves of polluting material in the same manner as do eastern oysters.

The requirements for sanitation within the shucking house and for proper conditions during shipment apply to Olympia oysters just as to eastern oysters, so far as the different methods of handling and shipping permit of such requirements. The oyster-shucking houses in and about Olympia and other northwestern cities are small and conditions can not be compared to those in the shucking houses of the Atlantic and Gulf coasts. The oysters are received in cloth sacks and are handled in comparatively small quantities.

The problems of green oysters and pink oysters have not yet arisen in the native Pacific coast industry. In fact, this small oyster industry of the Pacific coast has been comparatively free from many of the troublesome problems affecting the eastern Gulf coast industries. As the territory surrounding the oyster-producing regions becomes more thickly populated problems of pollution with sewage and trade wastes will be encountered but, profiting by the experience gained on the Atlantic coast, oyster growers and the State, Federal, and municipal authorities may take whatever action is necessary to preserve the industry.

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