Table II. Schedule for Peroxide Addition

Time of Addition in Hours	Ml. of 35% H_2O_2 Per 1000 Lb. of Albumen: Decreasing	Regular
0.00	200	216
0.25	100	72
0.50	90	72
0.75	84	72
1.00	76	72
1.25	70	72
1.50	64	72
1.75	59	72
2.00	54	72
2.25	50	72
2.50	45	72
2.75	42	72
3.00	38	72
3.25	35	72
3.50	32	72
3.75	30	72
4.00	28	72
4.25	25	72
4.50	23	72
4.75	21	72
5.00	20	72
5.25	18	72
5.50	16	72
5.75	15	72
6.00	14	72
6.25	13	72
6.50	12	72
6.75	11	72
7.00	10	72
TOTAL	1295	2232

Thus, equation 4 relates peroxide demand with time elapsed.

$$H = \frac{440\,y}{\text{antilog}\,\frac{T}{Z}}, \quad Z = \frac{(6.75)\,E}{75{,}000} \qquad (4)$$

Equation 4 may be used to calculate all additions except the one at zero time. This addition is twice the addition after the first time interval.

Table II contrasts the peroxide additions for the old and the new procedures, based on 15-minute intervals between additions, and 75,000 units of glucose oxidase per 1000 pounds of albumen.

Compare the total amounts of peroxide used in both procedures. In addition to eliminating foaming problems the decreasing addition schedule saves 40% of the peroxide.

The quantities of hydrogen peroxide calculated by the use of equation 4 are based on the almost complete exclusion of atmospheric oxygen. Depending on the vessel used and the type of agitation used, it may be possible to reduce this level somewhat.

For plant operations where a continued step-wise decrease in peroxide additions is not convenient, the author recommends the addition of 200 ml. of 35% hydrogen peroxide at zero time followed by, for the "eight-hour process," the addition of 100 ml. after or during each succeeding 15-minute interval through 2 hours, 50 ml. through 4 hours, 25 ml. through 6 hours, and 10 to 15 ml. to completion of the process.

Yolk or Whole Egg Stabilization

Yolk and whole egg can be desugarized by means of the glucose oxidase–catalase enzyme system too.

Since the pH of yolks is approximately 6.6, no pH adjustment is required. The yolks are warmed to 35 to 38° C., and two standard pounds of glucose oxidase per 1000 pounds of yolk are added. Peroxide is added at an over-all level of about 3 parts per thousand. This will reduce the glucose level from the initial 1% down to 0.1% dry basis in $3^1/_2$ hours. Increasing the enzyme level to 3 pounds per 1000 pounds will reduce the time to 2 hours.

Whole eggs contain about 1.2% glucose, dry basis. The pH of whole eggs is about 7.5. The process may be started here or, if desired, the pH could be lowered to 7.0 to 7.3. On whole eggs the general practice is to use 3 pounds of standard enzyme mixture to each 1000 pounds of whole eggs. The glucose level will be reduced to 0.1% dry basis in about 4 hours, with about $3^1/_2$ pounds of 35% hydrogen peroxide being added per 1000 pounds of whole egg.

As with the albumen, the process is rather flexible and may be modified to produce specialty products. The most common modification of enzyme-stabilized whole egg solids is to incorporate about 10% sucrose in the liquid whole egg prior to drying. The sucrose, being nonreducing, does not exert a deleterious effect on the resultant egg solids.

The use of the glucose oxidase–catalase system for the stabilization of foods subject to nonenzymatic browning is being developed. Application will no doubt be made to other products such as dehydrated mashed potatoes.

Evaluation of Method

It is difficult to evaluate any new development so soon after it has come out. Certainly no single development can solve all of the problems in the food field. There is hardly a development that can be made to work economically wherever theory says it should work. But oxygen and glucose are such basic causes for difficulty to the food technologist that we can confidently expect the commercial realization of this enzyme system to solve some of the commonest difficulties confronting the food technologist today.

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FOODS EVALUATION

Sampling Plan for Nutrition Research Program On Frozen Fruits, Juices, and Vegetables

H. P. SCHMITT, Research & Standards Department, National Association of Frozen Food Packers, Washington 5, D. C., and
R. J. JESSEN, Statistical Laboratory, Iowa State College, Ames, Iowa

A LITERATURE SEARCH of *Chemical Abstracts* over the past three and one-half years revealed some 20 papers dealing with the occurrence of one or more nutrients in fruits, juices, and vegetables after preservation by freezing followed by subsequent storage and/or cooking studies. In consideration of the great scarcity of such data, the few tabulations on the composition of raw and processed foods that include frozen foods may be open to question, especially

A sampling plan is presented for a program of research to determine the nutritional values (21 values for 700 sets of samples) from about 30,000 frozen food packages to be drawn by statistical procedure from commercial production of 48 frozen fruits, juices, vegetables during the 1953 and 1954 packing seasons in 149 plants throughout the nation.

in those circumstances that embody older data determined by less refined methods of analysis. For example, Irwin and Schuck (*2*) found that sodium values measured by older methods were higher than those determined by flame photometry. Dietitians, governmental agencies, home economists, homemakers, medical people, students, etc., have been actively searching for a complete and authentic reference on nutritional values for frozen foods—a reference that is at present nonexistent.

As a public service in recognition of the great need for this information, the National Association of Frozen Food Packers (N.A.F.F.P.) is sponsoring a five-year nutrition research program. Supporting funds have been subscribed by allied industry and by the association. The over-all program is being managed by a Nutrition Administrative Committee. A Research Subcommittee is responsible for the planning and coordination of research activities with the helpful counsel of nutrition scientists.

During the first stage of the program, the Wisconsin Alumni Research Foundation will determine approximately 15,000 nutritional values from some 30,000 frozen food packages to be drawn by statistical procedure from commercial production of 48 frozen fruits, juices, and vegetables during the 1953 and 1954 packing seasons in 149 plants throughout the nation. Data will be published in a series of papers listing values for ascorbic acid, β-carotene, folic acid, niacin, pantothenic acid, riboflavin, thiamine, vitamin B_6, calcium, magnesium, phosphorus, potassium, sodium, total iron, ash, caloric value (calculated from specific factors of the Atwater system), carbohydrate (by difference), crude fiber, fat (ether extract), moisture, and protein.

In later stages of the program, studies will be undertaken with the ultimate purpose of developing handling methods which will provide for maximum retention of nutrients during processing, storage, simulated commercial distribution, and cooking. These projects and restricted surveys on amino acids, choline, and iodine are expected to get under way in 1955 at universities well recognized in the science of nutrition.

Sampling Procedure

A study of the entire population of frozen food packages would be impossible and certainly impractical. However, modern methods of sampling are available so that the characteristics of a population can be easily and cheaply inferred from small samples. When these samples are drawn by some random procedure certain laws of probability are applicable. The probability laws permit one to determine the precision of the sample from the sample itself. Since an estimate is useless unless its precision limits are known, it is important that a random method of sampling the universe of frozen food packages be employed.

General information about the universe of frozen food packages to be sampled indicates that it is heterogeneous. Some of the principal sources of variability include region of production, climatic conditions, varietal differences, processing operations, style of pack, and method of packaging and freezing. Any efficient sampling program of such a heterogeneous population almost invariably requires stratification to increase the accuracy of the sample results.

It appeared reasonable to expect that the variation within plants is less than that among plants in the same region. Similarly, it appeared reasonable to expect that the variation among plants in the same region is less than that among plants in different regions. As a consequence of these expectations, a sample design was adopted such that regional differences were controlled through the use of stratification, and plant differences were met by obtaining the sample packages from many plants. It was decided to draw sample packages independently for each of the 48 frozen foods but to use a single basic plan for doing so.

Regional Sampling Statistically (*1*), the most efficient distribution of samples among the regions occurs when the number of packages sampled per region is made proportional to the product of the standard deviation of the character under measurement and the production of each region. When the standard deviation is an unknown value as in this case, it is then usually advisable to allocate the sample packages of a given product required from each region according to the production of that region. Using pack statistics data, it became a simple matter to allocate regional samplings according to production. Next, the regions were further subdivided into subregions, using the same pack statistics data for a given product by the procedure just described. Thus, the ultimate stratum was a geographical area of production from which a minimum of two sets of sample packages were selected. This area, that is stratum, was a region, subregion, or, in some cases, a group of regions. This technique takes advantage of known production to establish strata that are proportional to regional production. It restricts the randomness to a selection of plants within each stratum and thereby reduces sampling error to that existing within strata.

In instances of extremely large productions by a given plant, and in others where the number of primary packers within a region and/or stratum was very limited, it was necessary to deviate from the general plan of sampling directly proportional to production. Where such deviations were made, the exact proportions sampled are known and will be used in the analysis of the sample results. On all occasions, the sampling conformed as closely as possible to statistical requirements, and it will provide an objective estimate of means and their variances. Each stratum was assigned at least two samplings so that the mean, maximum, minimum, standard deviation, coefficient of variability, and standard error can be determined ultimately.

Scale of Sampling When the scale of sampling is increased, the accuracy of the estimate is increased for two reasons: (1) It permits a greater degree of stratification, and (2) it allows the larger sample size. Thus, it would appear advantageous to draw many samples for each of the 48 products being sampled in the first stage of the program. However, due to over-all limits on resources, it was necessary to keep the total amount of material sampled at a relatively fixed amount. Since each of the 48 products differs in degree of importance, it would not be wise to allocate the limited supply resources equally to each product. Large differences in volume of pack are consistently evident among the products listed in pack statistics. These data on volume of pack, consequently, provided a desirable and objective means of establishing the importance of each product for stage one of this program. Only pack statistics on retail sizes were used in evaluating the order of importance, since the primary purpose of the first stage is to establish the nutritional composition of frozen foods that the packer makes available to the homemaker.

Unit of Sampling The fundamental unit of sampling was a set of samples consisting of about 40 consumer-size packages of 25-ounce capacity or less, except for samples of frozen sliced apples and frozen apricots, where a set consisted of about ten, 5- to 10-pound packages, the smallest sizes produced. Each set of samples was drawn entirely from a given plant's production of a given product throughout the entire processing season.

The national sampling of each product was established on the following appraisal of the average national pack in retail-size packages during 1951:

One set of samples per million pounds of production for all products whose average volume ranged from an arbitrary minimum of 5,000,000 pounds to an arbitrary maximum of 15,000,000 pounds. This rate of sampling is equivalent to one sample package per 40,000 units of production in 10-ounce containers.

One set of samples was added to the established maximum for every 4,000,000 pounds packed in excess of the upper limit for all products, except for frozen concentrated orange juice, where an additional set was added for every 16,000,000 pounds beyond the maximum.

Table I illustrates the national sampling of products during 1953. Column one presents an alphabetical sequence of the 48 products. Column two lists the number of sets of samples calculated on the above basis. Column three gives the number of sets of samples required after an adjustment was made in consideration of (1) the estimated volume of pack, by style; and (2) instances of limited number of primary packers. For example:

(1) Fourteen sets were calculated for frozen asparagus, but since a ratio of two sets of asparagus spears to one of cuts and tips of asparagus was wanted in order to reflect the estimated production ratio of these styles of pack, an adjustment to 15 sets was made; and

(2) Thirty-two sets were calculated in the case of frozen concentrated orange juice. However, since there were only 22 primary packers, the total sets were reduced to 22 so that only one set was required from each plant.

Column four lists the number of sets of samples that packers have committed from their 1953 pack. Column five shows the number of plants being sampled for each product.

The geographical distribution of plants participating in the program is shown by Figure 1.

Example of Sampling The nation was divided into four regions: the Midwest, Northeast, South, and West, which are identical to those used by N.A.F.F.P. in compiling pack statistics. The sets of samples required for each product (column three of Table I) were allocated among these regions by a percentage appraisal of regional production of each product, and the sets required within a stratum were selected at random from the entire production of each product in the stratum. An example follows for the 24 sets of frozen sliced strawberries to be procured from the 1953 pack. Actual production figures, by plants and some states, are strictly confidential. Therefore, it is necessary to employ fictitious data on production in the example in Tables II, III, and IV.

Since each stratum must furnish at least two sets of samples, the Midwest and Northeast regions were combined to form one stratum. The South remained as a stratum because it was impossible to establish two strata supplying the required two and three sets of samples, respectively, and yet consisting of areas of production in relatively close geographical proximity. Considering the intra-

Table I. National Sampling of Products During 1953

Frozen Product	Sets of Samples: Calculated	Sets of Samples: Adjusted	Sets of Samples: Committed	Plants Sampled
Apples, sliced	5	5	6	6
Apricots	5	5	4	4
Asparagus	14	...	...	...
Cuts and tips	...	5	5	4
Spears	...	10	10	9
Beans	24	...	...	...
Cut green	...	12	11	10
French style	...	12	11	9
Beans, cut wax	5	5	5	3
Beans, Fordhook lima	23	23	23	19
Beans, thin-seeded lima	18	18	18	14
Blueberries	10	...	...	...
Cultivated	...	5	3	3
Wild	...	5	2	2
Boysenberries	5	5	2	2
Broccoli	21	...	...	...
Chopped	...	7	7	7
Spears	...	14	15	15
Brussels sprouts	16	16	16	12
Cauliflower	17	17	17	16
Cherries, R.S.P.	5	5	4	4
Collard greens, leaf or chopped	5	5	3	3
Corn, cut	15	15	16	16
Corn on cob	9	9	9	8
Grapefruit juice, concentrated	5	5	5[a]	5[a]
Grapefruit-orange juice blend, concentrated	5	5	5[a]	5[a]
Grape juice, concentrated	5	5	4	3
Kale, leaf or chopped	5	5	5	5
Lemonade, concentrated	19	9	6	5
Lemon juice, single strength	5	5	2	2
Mixed vegetables	17	17	18	15
Mustard greens, leaf or chopped	5	5	3	3
Okra	5	5	5	5
Orange juice, concentrated	32	22	22[a]	22[a]
Peaches, sliced	13	13	12	11
Peas, black-eye	5	5	5	5
Peas and carrots	12	12	13	12
Peas, green sweet	41	41	40	32
Pineapple, chunks	5	5	2	1
Pineapple juice, concentrated	5	5	2	1
Potatoes, diced for hash browning	5	5	3	3
Potatoes, French-fried	16	10	10	10
Potatoes, mashed or whipped	5	5	3	3
Raspberries, red	7	7	7	7
Rhubarb	5	5	5	5
Spinach	30	...	...	...
Chopped	...	15	4	3
Leaf	...	15	5	4
Squash, winter	5	5	4	3
Strawberries	29	...	...	...
Sliced	...	24	19	18
Whole	...	6	4	2
Succotash	7	7	7	7
Turnip greens, leaf or chopped	5	5	5	5
Total	495	471	412	368

[a] Includes samplings to be arranged in late 1953.

Table II. Distribution of 24 Sets of Frozen Sliced Strawberries Among Four Regions of Production

Region	Production: Millions of lb.	Production: %	Sets of Samples	No. of Strata
Midwest	2.56	3.6	1	1 (Midwest and Northeast)
Northeast	3.14	4.4	1	
South	15.00	21.2	5	1
West	50.08	70.8	17	3
Total	70.78	100.0	24	5

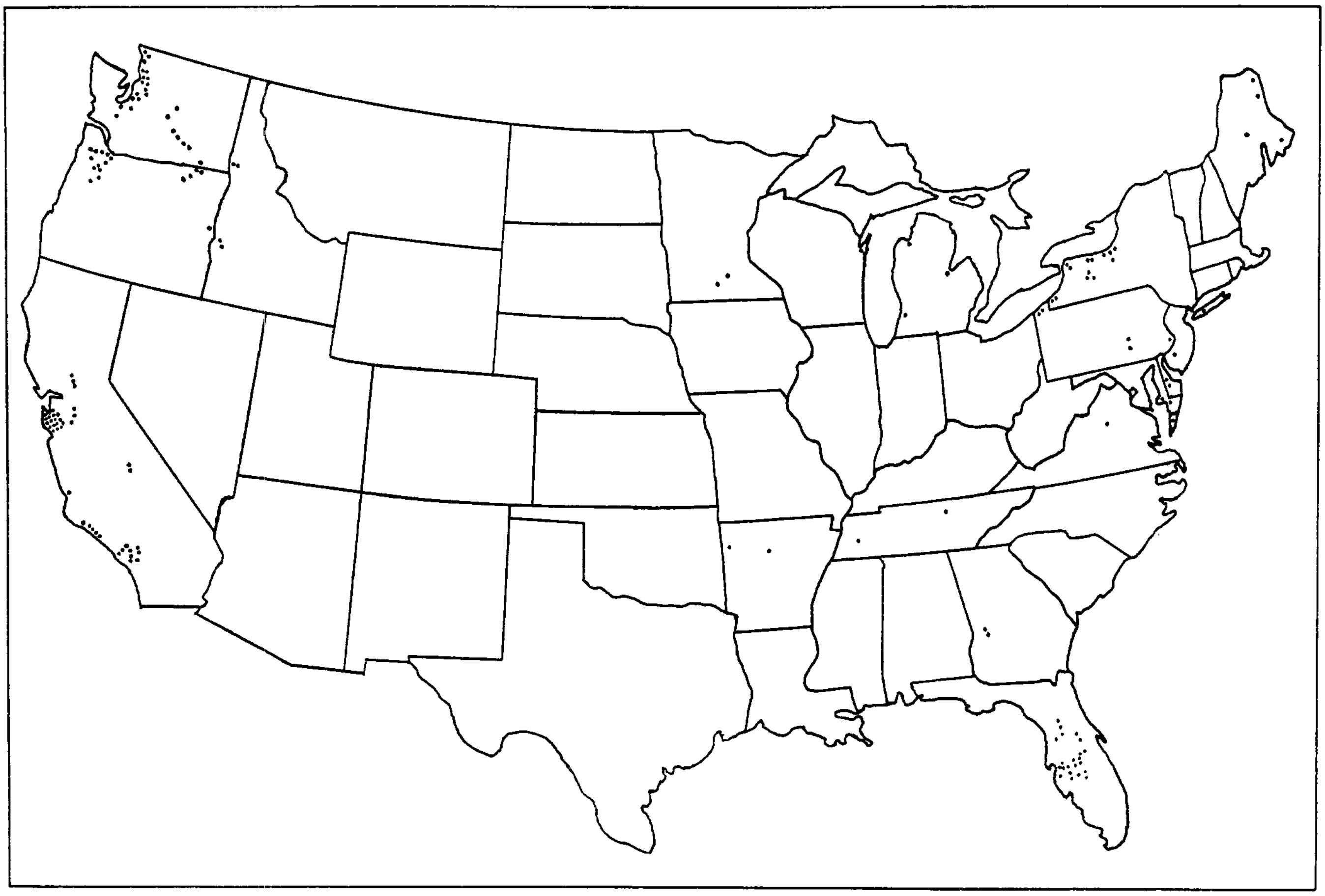

Figure 1. Approximate geographical location of plants participating in the program

regional distribution of production, the West was subdivided into three strata.

Frozen sliced strawberries were packed in only five states within the two-region stratum—Midwest-Northeast. These states and their respective production are given in Table III. The first step in random selection requires the computation of cumulative production, as shown by column three of Table III. In this example, two random numbers that fall between 0.01 and 5.70 must be found. Procedure for finding these numbers is to read down any three columns of numbers in a table of random numbers until sufficient numbers that fall within the required limits are observed. Decimals are mentally inserted in this operation. Returning to the example on the left, the last three columns of random numbers were used proceeding downward. The first number, 8.43, is rejected because it falls beyond the limits: 0.01 to 5.70. Since the second number, 3.83, falls within the limits, it is accepted. Likewise, 9.87 and 9.13 are passed over and 1.01 is accepted.

Random Numbers
09843
37383
44987
04913
24101
65702
27731
56811
35569

Rather than selecting two out of the five states for the sets of samples with equal probability, the selection has been purposively arranged so that each state has a different chance of being selected. Thus, all random numbers from 0.01 to 0.91 designate Indiana, all random numbers from 0.92 to 2.46 designate Michigan, etc. Hence, Indiana has a 0.91/5.70 chance of selection, whereas New York has a 1.65/5.70 chance, or nearly twice that of Indiana. This is the fundamental objective, because New York is the larger producer, and therefore, the sample will be more representative of production.

Since the random numbers, 3.83 and 1.01, were the first two eligible numbers in this example, it can be seen as a consequence that Michigan and New York were selected to furnish the two sets of samples. The next step was to determine which of several plants in each of the selected states would be sampled.

Table III. Random Selection of Two States to Be Sampled in Midwest-Northeast Stratum

States	*Production, Millions of Lb.* — *Actual*	*Production, Millions of Lb.* — *Cumulative*	*Sets to Furnish*
Indiana	0.91	0.91	0
Michigan	1.55	2.46	1
Minnesota	0.09	2.55	0
New York	1.65	4.20	1
Pennsylvania	1.50	5.70	0
Total	5.70		2

After listing the New York plants and their respective volume, the order of which is immaterial, plant h was selected by a similar application of the above procedure. This is shown by Table IV. One plant was similarly selected in Michigan. These two plants were contacted to supply the required sets of samples for the Northeast-Midwest stratum.

A similar procedure was followed in the other regions. Random selection of the five sets of samples required from the South, which in this case consisted of a single stratum, placed two sets with plants in Tennessee and one set with plants in Arkansas, Georgia, and Oklahoma. The West was subdivided into the three strata—California, Oregon, and Washington. Random selection within each of these respective strata placed seven sets with six plants in California, six sets with six plants in Oregon and four sets with four plants in Washington.

In order to achieve better coverage of interplant variation, the sampling of a given product from any one plant was limited to two sets in all instances where random selection brought a given plant into the sampling more than twice. Random selection of this type favors the sampling of large productions and yet opportunity exists for small plants to come into the sampling program in their

Table IV. Random Selection of a Plant in New York State to Furnish One Set of Samples on Sliced Strawberries

Plants	Production, Millions of Lb. Actual	Cumulative	Plant Selected
a	0.08	0.08	...
b	0.12	0.20	...
c	0.14	0.34	...
d	0.21	0.55	...
e	0.22	0.77	...
f	0.26	1.03	...
g	0.29	1.32	...
h	0.33	1.65	x
Total	1.65		

proportionate share. The packers thus selected to supply the samples will probably pack upward of 75% of the total production in 1953, and a realistic coverage of the many variants will be realized.

At-Plant Sampling

Copies of Form S-3, "Official Procedure for Drawing At-Plant Samples During 1953," (copies of this form and of others mentioned herein are available from N.A.F.F.P. and will be sent upon request) were sent to quality control supervisors and plant managers for their use in drawing samples by uniform procedure. A set of samples for testing will be saved for each product that a given plant furnishes. A companion set for reserve will also be saved in the event that the first set is lost or damaged in transit.

Each package within a set will be drawn from normal production within an hour of specified intervals of production throughout the entire processing season. Thus each set will include packages representing all grades of product, all types of retail sizes, plus intraseasonal and other intraplant variations, because the character of each package will depend only upon the type of product material being processed when the specified intervals of production are reached.

The intervals of production consist of an initial point of production when the first sample package is drawn and a series of consecutive and repeating intervals of production when successive sample packages are drawn. Each plant will save about 40 packages per set regardless of its total output. The intervals were determined by N.A.F.F.P. from pack statistics on retail production in 1952 and were sent to packers on Form S-4, "Strictly Confidential Sampling Rates." In some instances, it was more practical for the packer to employ Forms S4-b, S4-c, or S4-d, "Guide for Ascertaining Sampling Rates," to establish rates of sampling from his estimated production of a given product in retail sizes during 1953.

Presume that a plant packs 781,000 pounds of frozen sliced strawberries in retail sizes. The confidential sampling rate for this plant, as assigned by N.A.F.F.P., would consist of an initial sampling at an interval of production equal to 1000 pounds and a rate of sampling at successive intervals of production equal to every 20,000 pounds packed after the initial sampling. Thus, the first sample package of the set would be drawn within an hour of the time that 1000 pounds were packed, the second when 21,000 pounds were packed, the third when 41,000 pounds were packed, etc., throughout the balance of the season.

Products packed in cartons and metal-end cartons were sampled immediately after the package passed through either the overwrap or closing machine. Products packed in all-metal containers and frozen by immersion freezing were sampled by drawing the package from the batch or lot of cans frozen in the metal basket corresponding to the specified intervals of production. Products packed in all-metal containers and not frozen by immersion freezing were sampled after the can passed through the closing machine.

A pink, wrap-around label band bearing code and product name in indelible ink was attached to each sample package. The month, day, and hour of sampling were written in the appropriate space and the sample was returned to the line for a trip through the freezer. Personnel at the freezer station removed all packages bearing the pink labels before customary casing operations and placed them in a case or box bearing an identical label. All samples were held in storage at 0° F., or lower, until an N.A.F.F.P. representative called to collect the sets for testing.

Duplicate copies of Form S-5, "Average Seasonal Sample History," were completed for each set of samples for testing and for any sample package or packages drawn from abnormal or unusual processing operations. This information includes code and product, all varieties, data on time, type, and temperature of blanch, special treatment, quantity and type of optional ingredients, method and temperature of freezing, general product quality, storage temperature, ratio of fruit to packing medium, quantity and density of liquid packing medium, type of sweetening agent, and any additional comments the packer wished to make about the samples.

At the close of the season for each product, the packer will complete Form S-6a by writing in the number of packages in the set and the date the set was completed. This self-addressed postal card bearing code and product designation will then be returned to N.A.F.F.P. Form S-6b, "At-Plant Sample Inventory," is available for the packer's optional use in keeping a record of the sets of samples he furnishes for testing.

Shipping and Testing Samples

Members of the Nutrition Research Subcommittee and other interested parties will act as N.A.F.F.P. representatives to collect and ship the sets of samples for testing via Church Freight Service with Ryan recording thermometers to a refrigerated warehouse in Madison, Wis.

C. H. Krieger, L. J. Teply, and P. H. Derse of the Wisconsin Alumni Research Foundation in Madison, Wis., will be in charge of the vitamin, mineral, and proximate assays.

A representative slurry will be prepared from all packages within a set on an as is, uncooked basis. Each slurry will be subjected to all of the prescribed analyses, unless mutually agreed that a given assay shall be excluded. Such exclusion would be made because the nutritional component in question was found to be present in insignificant quantities after a minimum of three sets of available samples of a given product, or 25% of the sets of available samples of a given product, whichever is greater, have been tested. The number of sets used in determining significance will be reasonably spread across the four basic regions of production.

Summary

The statistical sampling plan presented is the basis for a series of research papers to follow, which will report data obtained from nutritional tests completed during the first stage of this research program. These papers will interpret approximately 15,000 determinations (21 values for some 700 sets of samples) on about 30,000 frozen food packages drawn by statistical procedure from commercial production of 48 frozen fruits, juices, and vegetables during the 1953 and 1954 packing seasons in 149 plants throughout the nation.

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