

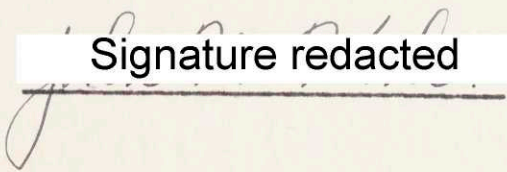
THE DEVELOPMENT OF TESTING METHODS FOR THE
EVALUATION OF WRAPPING MATERIALS
USED FOR FOOD PACKAGING.

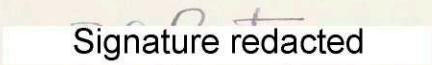
by

John M. Kohr

Submitted in partial fulfillment of the requirements
for degree of
Bachelor of Science
from the
Massachusetts Institute of Technology
1935
Course VII

Signature of Author,


Signature redacted


Signature redacted

384 Marlborough Street
Boston, Massachusetts
May 16, 1935

Professor B. E. Proctor
Massachusetts Institute of Technology
Cambridge, Massachusetts

Dear Sir:

I should like to submit this thesis for
consideration in the Sigma Xi Thesis Prize Contest.

Very truly yours,


Signature redacted

Mr. George W. Swett,
Secretary of the Faculty,
Massachusetts Institute of Technology.

Dear Sir:

I submit herewith my thesis in partial fulfillment of the requirements for the degree of Bachelor of Science from the Massachusetts Institute of Technology.

Very truly yours,


Signature redacted

ACKNOWLEDGMENTS.

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Mosinee Paper Mills Company

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Introduction.

The packaging of foods for protection against contamination during storage, handling, and exposure for sale has greatly increased in recent years, and has led to the development of numerous types of packaging materials. There are certain definite reasons for the use of almost any of the many types; some furnish protection from moisture gains or losses, and some from the spoilages caused by the entrance of light, oxygen, insects, and odors and flavors. Still other types are used because they are attractive, and others because they are transparent, thus placing the product on display.

Nearly all food products should be protected against spoilage which may be caused by one or more different conditions. It should then be the object to use, with any specific product, the wrapping material which is best adapted to the purpose. Since an attractive package is a very important point to be regarded, the package must be made of attractive materials and in an attractive manner, as well as have the properties requisite to the best possible protection of the product. Some of these types are transparent and semi-transparent materials, metal foils, parchments, glassines, and grease-proof materials.

A discussion of the methods of manufacture and the properties of these materials will not be undertaken because of the many different kinds considered, the widely different methods of manufacture used, and the different properties of

each. The uses of each type of packaging material may differ widely but in general it may be said that at least one type is being used for almost any food product.

The question arises as to whether or not the packaging material used for a specific product has all the necessary properties to keep the product in sound condition for long periods of time. It is doubtless true that certain products could be kept in better condition for a longer time if the package were made of materials having all the requisite properties. In some cases it is impossible to find all the necessary qualities of a single material. The situation could be helped a great deal if the properties of all the various materials were known. Then each material could be used where it is best suited.

There are many different properties to be considered, and the methods for determining many of them are inadequate from the standpoint of comparisons between the various materials. Consideration of some of these properties led to the adoption of this subject for my thesis.

Statement of the Problem.

Because there are no commonly accepted methods of determining some of the more important properties of food wrapping materials, the purpose of this thesis is to study those advocated and attempt to develop others which will make a suitable basis for the comparison of the properties of the various types. The first problem was to determine the most important properties for which testing methods were either lacking or inadequate.

After consideration of a group of food products which are commonly packaged, and the requirements of the package for keeping them in sound condition for long periods of time, the following properties were considered to be the most important:

The ability to resist the penetration of,

1. Moisture
2. Odors and Flavors
3. Air or oxygen
4. Microorganisms
5. Light

The purpose, then, is to determine suitable testing methods for these properties. The testing methods should be of such a nature that comparative determinations can be made, as well as roughly quantitative determinations.

The determination of testing methods for each of the above properties will be treated as a separate section in the pages which follow.

Section 1.

THE PENETRATION OF MOISTURE.

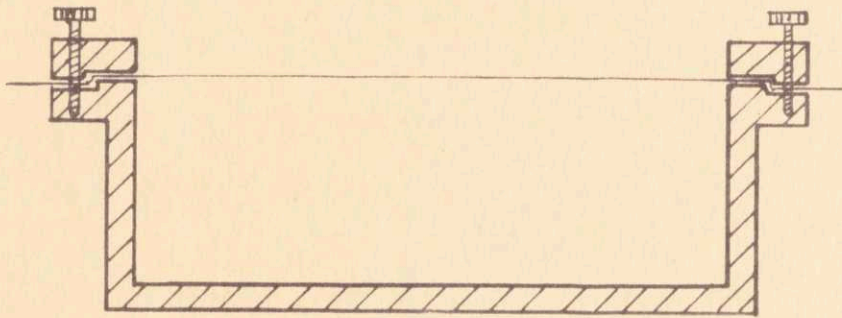
THE PENETRATION OF MOISTURE.

Review of the Literature.

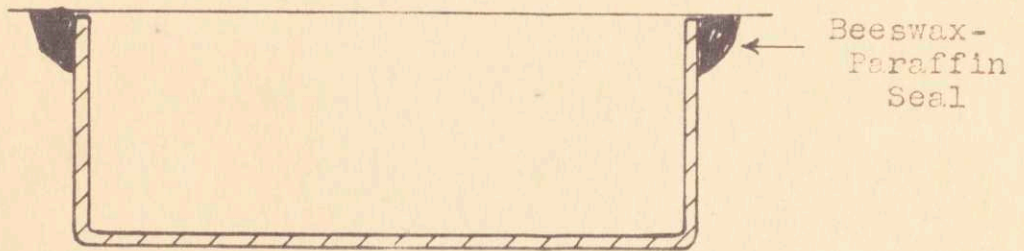
A very good method for the determination of the penetration of Moisture is the Abrams Cup Method. It consists of placing a cup covered with the sample in a constant temperature and humidity cabinet and measuring the loss in weight of the cup. The cup is made especially for the purpose and is pictured in the accompanying diagram, Fig. 1. It is constructed of aluminum and the principle of the seal can be seen from the step-like construction of the ring and the top of the cup. Water is placed in the cup and the loss in weight during the test period of seven days is determined. The cup is placed in a small cabinet in which the temperature is maintained at 70°F and the relative humidity at 50%. The air in the cabinet is circulated by means of a small fan so that at all times there is a differential in the humidity within the cup and in the cabinet of 50%. The above temperature and humidity differentials were chosen because they were believed to simulate actual conditions more closely than any other. The test period is seven days in length and the cups are weighed every day. This method is designed to measure the exact penetration of moisture through the sample and the penetration is expressed in grams/ 24 hours/ 100 sq. in./ 70°F./ 50% humidity. Some test results obtained by this method are as follows:

Open Cup	150	
Tissue	63	
Cellulose Acetate	40	
Glassine	30	
Veg. Parchment	25	
Dry Waxed	10	
Metal Foil	.4	(1)

Diagram No. 1.



Abram's Aluminum Cup



Crystallizing Dish

An improvement of this method is the one in use in the Frosted Foods Sales Corporation laboratory in Boston. In this method a three-inch crystallizing dish is used in place of the aluminum cup, Fig. 1. The sample is sealed on the dish with a mixture of 36% Beeswax and 64% Paraffin. This seems to be slightly better than the Abrams Method in that the seal between the sample and the cup does not allow so much leakage as does the seal in the case of the aluminum cup.

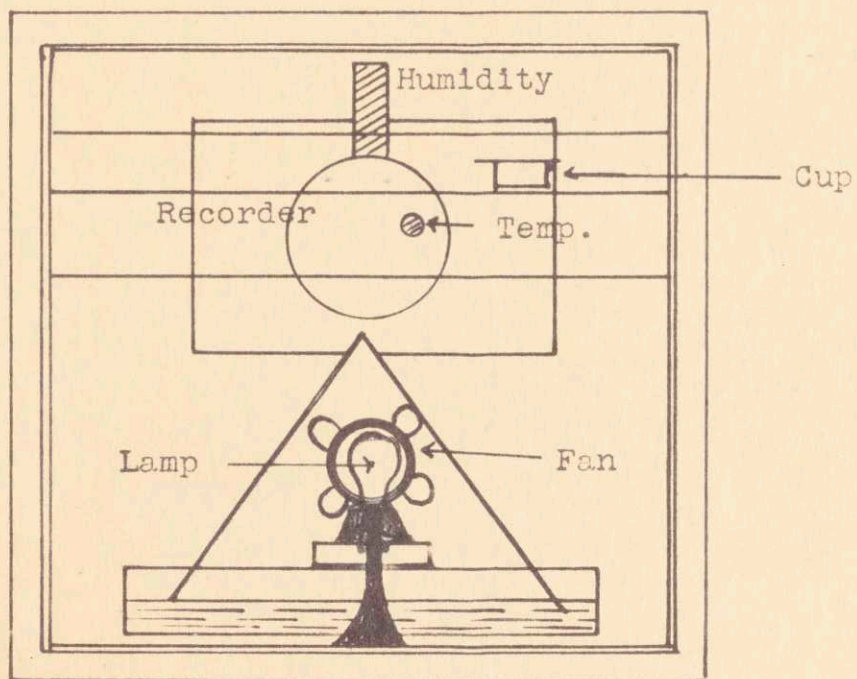
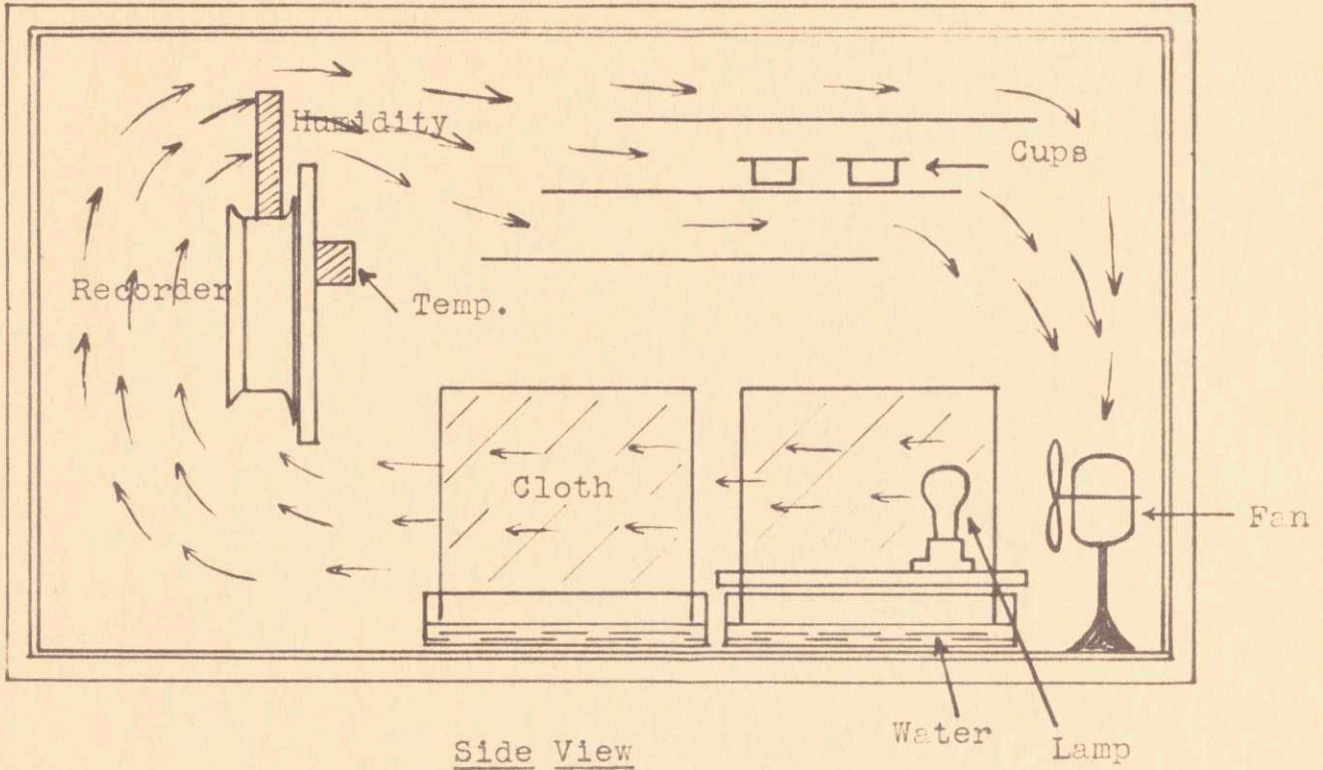
Experimental Work.

A method was tried embodying the use of both of these methods and at the same time making conditions a little more severe so that comparative results could be had in a shorter time. Under these more severe conditions greater amounts of moisture penetrate the sample.

The cup used was the same as that used by the Frosted Foods Sales Corporation and shown on the previous diagram, Fig. 1. The arrangement of the high humidity cabinet, constructed by the author and used in this method is shown on the accompanying diagram, Fig. 2. In place of water used in the cups in the previously described methods calcium chloride was used. The moisture transfer in this case was into the cup rather than out of it as in the previously described methods. The cups were placed in the high humidity cabinet operating at a temperature of 80°F with a relative humidity of from 97 to 99%. In this case the cups were weighed every day for three days and the average gain in weight was taken

Diagram No. 2.

Structure and Arrangement
of High Humidity Cabinet.



to be indicative of the moisture penetration through the sample.

The constant temperature was maintained by means of a sensitive thermostat which controlled the operation of an electric lamp used as a source of heat. The humidity was maintained as high as possible by allowing water to evaporate from large pieces of cloth dipping into pans of water. A stream of air was blown over the wet cloths in order to increase the evaporation rate and also to keep the conditions of temperature and humidity the same in all parts of the cabinet.

A constant record of the temperature and relative humidity was kept at all times by means of a Foxboro recording thermometer and hygrometer built in a single unit and recording on the same sheet.

The first difficulty encountered was that of keeping a tight seal between the sample and the cup. Under these rather severe conditions some of the samples would shrink to such an extent that the seal would be broken. At first paraffin alone was used as the sealing material but this was found to be entirely unsatisfactory. The use of a mixture, 35% beeswax and 65% paraffin, was found to be unsatisfactory also. Then a mixture using 50% beeswax and 50% paraffin was tried and found to be satisfactory under all conditions of the test.

At first 5 grams of calcium chloride were placed in the cups to absorb the moisture which penetrated the sample. This was found to absorb moisture more slowly toward the end of the three day test period than in the beginning. After

this was noticed, the amount of calcium chloride was increased to 8 grams. The following figures will show the purpose of this.

<u>Sample</u>	<u>Time</u>			<u>Amount of CaCl₂</u>
	<u>1 day</u>	<u>2 days</u>	<u>3 days</u>	
Parchment	4.810 g.	3.433 g.	2.975 g.	5 g.
The Same	5.170 g.	4.464 g.	4.044 g.	8 g.

Conditioning the samples of wrapping materials in the high humidity cabinet for a period of 24 hours previous to use was tried but it was found that many of the samples became so damp that it was impossible to seal them on the cups. For this reason the procedure of conditioning the samples was abandoned.

Several aluminum cups of the type used in the Abrams Cup Method were borrowed from Mr. Fitzgerald of the Frosted Foods Sales Corporation. These were tried in comparison with the method of sealing the sample on a three inch crystallizing dish with the mixture of beeswax and paraffin. In general it was found that a much greater penetration through the same area was had when using the aluminum cups. Since the samples were the same the greater penetration of moisture must have been caused by the different type of seal between the sample and the cup. This indicated that the seal was not tight so their use was discontinued.

<u>Type of Cup</u>	<u>Time.</u>		
	<u>1 day</u>	<u>2 days</u>	<u>3 days</u>
Aluminum	.123 g.	.095 g.	.094 g.
Three-inch cryst. dish	.078 g.	.047 g.	.039 g.

Summary and Conclusions:

As a result of these experiments the most satisfactory method was that using three-inch crystallizing dishes containing 8 grams of calcium chloride, the sample being sealed on the dish with a mixture of 50% Beeswax and 50% Paraffin. These dishes were placed in a high humidity cabinet operating constantly at a temperature of 80°F. and with a relative humidity of from 97 to 99%. For the measurement of the penetration of moisture, these dishes were weighed each day for a three day test period, and the average gain per day computed. This figure gives the amount of moisture which penetrated a circular sample three inches in diameter or having an area of 7.07 square inches.

The types of wrapping material which proved to be the best from the standpoint of moisture penetration were the metal foils, next to these came the rubberized materials and the Cellophane type materials.

A complete compilation of all the test results obtained using this method follows.

<u>Sample</u>		Weight in grams								
		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>	
Control	1	56.437	56.492	.055	56.524	.032	56.554	.030	.039	
	2	50.058	50.104	.046	50.127	.023	50.159	.032	.034	
Parchment Type	3	65.866	Seal Broken							
"	"	4	52.797	Seal Broken						
"	"	5	46.487	51.297	4.810	55.730	3.433	58.705	2.975	3.706
"	"	6	49.614	49.643	.029	49.671	.028	49.699	.028	.028
Glassine Type	1	50.238	50.403	.165	50.555	.152	50.674	.119	.145	
"	"	2	54.467	58.913	4.446	63.419	4.516	66.622	3.203	4.052
"	"	3	59.740	64.530	4.790	Seal Broken				
Paper		47.237	51.646	4.409	56.381	4.735	60.033	3.652	4.265	
Cellophane Type	1	56.437	56.492	.055	56.524	.032	56.554	.030	.039	
"	"	2	54.003	Seal Broken						
"	"	3	50.058	50.104	.046	50.127	.023	50.159	.032	.034
"	"	4	58.314	Seal Broken						

Penetration of Moisture. Group 1.

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- Straight Paraffin
 Absorbent Material -- 5 g. Calcium Chloride

<u>Sample</u>		<u>Weight in grams.</u>							
		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>
Control	1	57.313	57.359	.046	57.403	.044	57.446	.043	.044
	2	67.786	67.836	.050	67.902	.066	67.948	.046	.054
Parchment Type	3	57.555	64.171	6.616	69.135	4.964	74.557	5.422	5.664
" "	4	55.445	62.035	6.590	66.830	4.795	70.812	3.982	5.122
" "	5	47.596	52.766	5.170	57.230	4.464	61.274	4.044	4.559
" "	6	53.244	53.264	.020	53.286	.022	53.311	.025	.022
Glassine Type	1	52.069	52.151	.082	52.222	.071	52.308	.086	.080
" "	2	59.750	65.399	5.649	69.985	4.586	74.255	4.270	4.835
" "	3	54.172	59.959	4.787	64.873	4.914	69.287	4.414	5.038
Cellophane Type	1	57.313	57.359	.046	57.403	.044	57.446	.043	.044
" "	2	58.818	Seal Broken						
" "	3	67.786	67.836	.050	67.902	.066	67.948	.046	.054
" "	4	60.212	Seal Broken						
Metal Foil	1	49.816	49.844	.028	49.872	.028	49.902	.030	.029

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- 35% Beeswax and 65% Paraffin
 Absorbent Material -- 8 g. Calcium Chloride

Penetration of Moisture. Group 2.

<u>Sample</u>		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>Weight in grams.</u>			<u>Gain</u>	<u>Average Gain</u>	
					<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>			
Control	1	70.524	70.572	.048	70.592	.020	70.666	.054	.034	
	2	52.894	52.910	.016	52.937	.027	52.963	.026	.023	
Glassine Type	4	54.198	Seal Broken							
"	"	5	56.412	Seal Broken						
"	"	6	61.354	67.254	5.900	72.418	5.164	77.987	5.569	5.511
"	"	7	57.539	Seal Broken						
"	"	8	59.632	64.750	5.118	69.947	5.197	75.191	5.244	5.186
"	"	9	48.534	Seal Broken						
Parchment Type	1	59.929	66.019	6.090	70.749	4.730	74.774	4.025	4.615	
Cellophane Type	1	70.524	70.512	.048	70.592	.020	70.666	.054	.034	
"	"	2	67.812	Seal Broken						
"	"	3	52.894	52.910	.016	52.937	.027	52.963	.026	.023
"	"	4	54.706	Seal Broken						

Penetration of Moisture. Group 3.

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- 50% Beeswax and 50% Paraffin
 Absorbent Material -- 8 g. Calcium Chloride
 Samples conditioned in incubator for 24 Hrs.

Weight in grams.

<u>Sample</u>		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>	
Control	1	51.826	51.894	.078	51.941	.047	51.970	.039	.054	
Glassine Type	4	56.102	56.162	.060	56.206	.044	56.240	.034	.046	
"	"	5	54.309	54.382	.073	54.416	.034	54.446	.030	.046
"	"	6	59.081	65.256	6.175	69.975	4.719	73.446	3.471	4.788
"	"	7	52.305	52.620	.315	52.864	.244	53.111	.247	.235
"	"	8	58.953	63.533	5.580	68.405	4.872	71.920	3.515	4.656
"	"	9	53.263	53.502	.239	53.619	.117	53.784	.165	.140
Parchment Type	1	61.767	67.474	5.707	72.585	5.111	76.489	3.904	4.907	
"	"	2	48.507	54.003	6.096	59.589	4.987	63.317	3.728	4.933
Cellophane Type	1	51.826	51.894	.078	51.941	.047	51.970	.039	.054	
"	"	2	67.250	73.392	6.142	78.134	4.742	81.720	3.586	4.823
"	"	4	56.263	62.002	5.739	66.817	4.815	70.259	3.442	4.662

Penetration of Moisture. Group 4.

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- 50% Beeswax and 50% Paraffin
 Absorbent Material -- 8 g. Calcium Chloride

<u>Sample</u>		<u>Weight in grams.</u>								
		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>	
Control	1	53.111	53.197	.086	53.246	.049	53.288	.062	.072	
	2	53.990	54.045	.055	54.071	.026	54.091	.020	.034	
Glassine Type	10	48.684	54.838	6.154	60.058	5.220	64.239	4.181	5.185	
"	"	11	61.332	67.096	5.764	70.039	2.943	75.834	5.795	4.834
"	"	12	52.651	57.742	5.091	62.680	4.938	66.504	3.824	4.617
"	"	13	58.217	63.670	5.453	68.707	5.037	73.140	4.433	4.974
"	"	14	57.086	51.781	4.695	66.281	4.500	70.098	3.817	4.334
"	"	15	55.145	55.592	.447	56.006	.414	56.430	.424	.428
Rubberized	3	60.047	60.074	.027	60.096	.022	60.121	.025	.025	

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- 50% Beeswax and 50% Parrafin
 Absorbent Material -- 8 g. Calcium Chloride

Penetration of Moisture. Group 5.

Weight in grams.

<u>Sample</u>		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>	
Control	1	51.913	51.970	.057	52.005	.035	52.035	.030	.041	
	2	60.463	60.542	.079	60.580	.038	60.613	.033	.050	
Metal Foil	1	50.750	50.776	.026	50.787	.011	50.812	.025	.021	
"	"	2	52.854	52.854	.000	52.859	.005	52.860	.001	.002
"	"	3	56.807	56.807	.000	56.807	.000	56.807	.000	.000
"	"	4	59.275	59.275	.000	59.276	.001	59.278	.002	.001
"	"	5	47.210	47.210	.000	47.210	.000	47.212	.002	.001
"	"	6	68.848	68.848	.000	68.851	.003	68.854	.003	.002
"	"	7	60.551	60.551	.000	60.551	.000	60.551	.000	.000

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- 50% Beeswax and 50% Paraffin
 Absorbent Material -- 8 g. Calcium Chloride

Penetration of Moisture. Group 6.

GROUP 7.

Weight in Grams.

<u>Sample</u>		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>
Control	1	68.931	69.017	.086	69.040	.023	69.085	.045	.051
	2	52.373	52.464	.091	52.492	.028	52.560	.068	.062
Rubberized	1	58.535	58.557	.022	58.576	.019	58.596	.020	.020
	2	55.086	55.102	.016	55.115	.013	55.128	.013	.014

Incubator at 80°F and 97 - 99% Relative Humidity
 Sealing Material -- 50% Beeswax and 50% Paraffin
 Absorbent Material -- 8 g. Calcium Chloride.

GROUP 8.

<u>Sample</u>		<u>Start</u>	<u>24 Hrs.</u>	<u>Gain</u>	<u>48 Hrs.</u>	<u>Gain</u>	<u>72 Hrs.</u>	<u>Gain</u>	<u>Average Gain</u>
Control	1	56.437	56.492	.055	56.524	.032	56.554	.030	.039
Cellophane Type 1		110.176	110.299	.123	110.394	.095	110.488	.094	.104
Parchment Type 6		114.151	114.256	.105	114.310	.054	114.364	.054	.071

Incubator at 80°F and 97 - 99% Relative Humidity
 Absorbent Material -- 8 g. Calcium Chloride.
 Used Abrams Method Aluminum Cups

Penetration of Moisture. Groups 7 and 8.

Section 2.

THE PENETRATION OF ODORS AND FLAVORS.

THE PENETRATION OF ODORS AND FLAVORS.

Review of Literature.

As far as could be determined from the literature there was no work done previously on a method of determining the relative amounts of odors and flavors, which penetrate food wrapping materials, using any other criteria than the organoleptic tests. The inadequacy of such methods is obvious as not all persons who may have occasion to perform similar tests have the same sensitivity of smell. This would lead to results which could not be compared in any way. Odors and flavors are very closely allied so that in many cases the odor of some volatile constituent in a food product is entirely responsible for the flavor of that product.

Experimental Work.

It was decided to measure quantitatively the amount of some volatile material which could penetrate the sample. The following method of measuring the amount of ammonia which would penetrate was considered but not used. Place some ammonia-free water in a three-inch crystallizing dish and seal the sample firmly over the top of the dish. The dish should then be placed in a small desiccator in which some concentrated ammonium hydroxide had previously been placed. By allowing the apparatus to stand in this condition for several hours some of the ammonia would certainly pass through the sample and be absorbed by the ammonia-free water in the crystallizing dish. The amount of ammonia could then be determined by

Nestlerizing the sample of water and comparing the color obtained with that of a standard in a colorimeter. This system was not tried however, because ammonia has a relatively small molecule and it was believed that it would be exceedingly hard to secure a sample of wrapping material which would not allow the passage of large amounts in a very short time. A modification of this plan was adopted for trial after further consideration.

Most of the compounds which are responsible for odors and flavors are organic in nature and have relatively large molecules. In order to have a test resembling natural conditions a compound of this nature should be used.

A list of a large number of essential oils of fruits was obtained from Mr. Fitzgerald of the Frosted Foods Sales Corporation of Boston. Of this group of odoriferous materials, one of the aldehydes, tolyl aldehyde (methyl-benzyl aldehyde), was chosen. The Fuchsine-Sulphurous Acid Test was tried with this aldehyde and found to be very delicate. (The solution will respond to formaldehyde 1 ; 10,000 with 5 minutes.)

The following test solution was used:

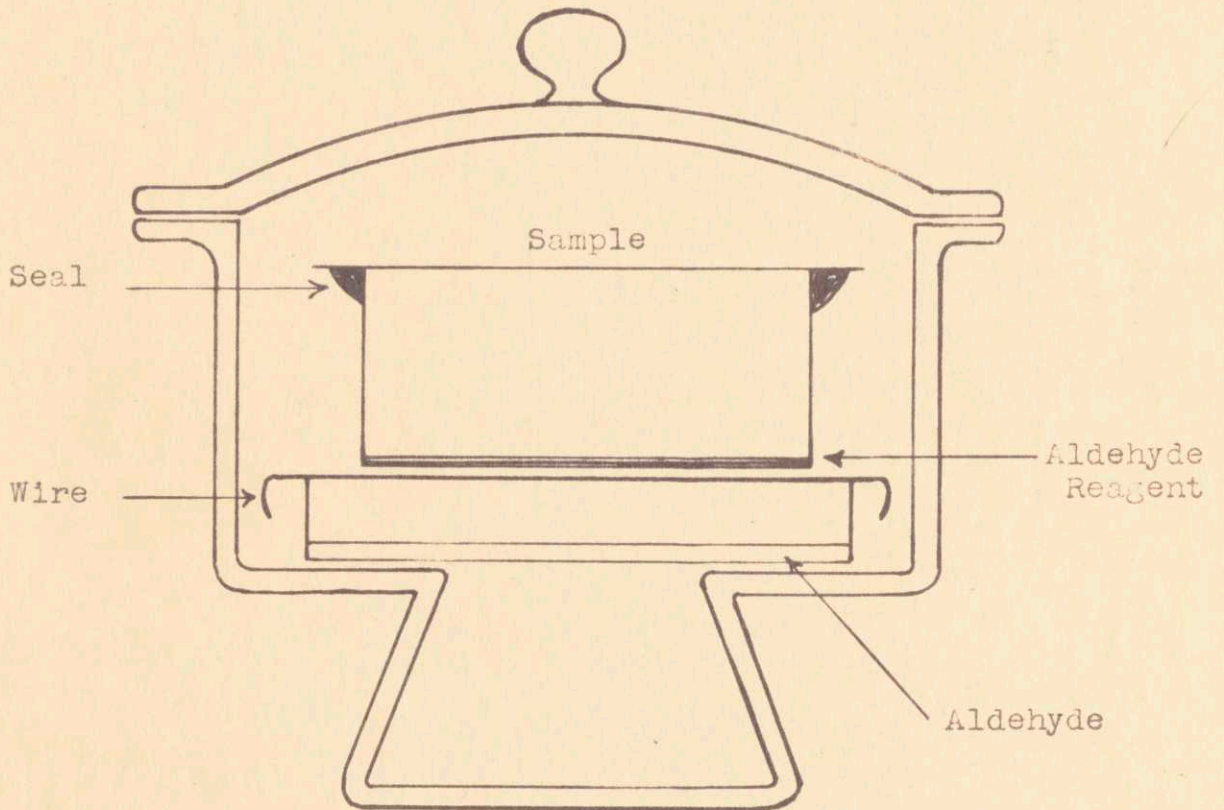
Fuchsine-Sulphurous Acid Test Solution. Dissolve 0.2 g. fuchsine in 120 ml. hot distilled water, cool the solution, add solution of 2 g. anhydrous sodium sulphite in 20 ml. of distilled water, and follow by 2 ml. of concentrated hydrochloric acid. Then dilute the solution to 200 ml. and allow to stand for one hour before use. (2)

This solution may be kept in perfectly good condition in small amber bottles for two months without loss of its sensitivity.

The question then arose as to whether the red coloration of the fuchsine-sulphurous acid solution would take place in the presence of the vapors of the aldehyde. The aldehyde was

Diagram No. 3.

Apparatus for Measuring Penetration
of Odors and Flavors.



placed in the bottom of a small desiccator and 5 ml. of the reagent, in a small dish, were placed in the desiccator. It was observed that a faint red coloration appeared within fifteen minutes, and within two hours this had deepened to almost purple.

The same procedure was then tried placing 5 ml. of the aldehyde reagent in a three-inch crystallizing dish with a sample of wrapping material sealed over the top. The dish was then placed in the desiccator. With a sample sealed over the dish the time required for the reagent to change color was from four to seven hours. It was decided to use the latter time, seven hours, as the test period.

In order to determine the relative amounts of aldehyde which penetrated the sample, the color of the reagent was compared with a standard in a colorimeter. The standard was prepared by adding 0.01 ml. of the aldehyde to 5 ml. of the fuchsine-aldehyde reagent.

Summary and Conclusions.

The test method which was found to give comparable results for the measurement of the penetration of odors and flavors was that of placing the odoriferous substance, methylbenzyl aldehyde in this case, in the bottom of a small desiccator, and placing fuchsine-sulphurous acid reagent in a three-inch crystallizing dish covered with the sample of material to be tested. The test period found to work well was seven hours. At the end of this time the color of the aldehyde reagent was compared with the color of a standard in a Bausch and Lomb

colorimeter. The standard was prepared by adding 0.01 ml. of the aldehyde to 5 ml. of fuchsine-sulphurous acid reagent. By means of this comparison of colors it was possible to determine the amounts of aldehyde which penetrated the sample during the seven hour test period. This amount is the amount of aldehyde which penetrated a circular sample three inches in diameter or 7.07 square inches.

The types of wrapping material which was best from the standpoint of odor and flavor penetration were the metal foils, next to these came the rubberized materials and the Cellophane type materials.

A complete compilation of the test results obtained using this method follows.

Recommendations.

This method makes it possible to measure the penetration of a single type of odoriferous material and as a continuation of the work methods should be devised to measure the penetration of other types of odor and flavor producing substances.

Longer times of exposure of the sample to the tolyl aldehyde should also be tried so that those which showed no coloration would show some evidence of the penetration of the odoriferous material.

Penetration of Odors and Flavors.

<u>Sample</u>	<u>Exposed 7 Hours.</u>		Corrected for standard reading 1.0.
	Std.	Test	
Cellophane Type 1	No Coloration		No Color
" " 2	3.1	4.3	1.4
" " 2	3.0	4.0	1.3
" " 3	No Coloration		No Color
" " 4	.7	9.9	14.1
" " 4	.7	9.1	12.9
Parchment Type 3	1.1	12.1	11.0
" " 4	1.1	8.7	7.9
" " 5	.2	15.0	7.5
" " 6	No Coloration		No Color
Paper Type	1.1	13.0	11.8
Rubberized Type 1	No Coloration		No Color.
" " 2	"	"	" "
" " 3	"	"	" "
Metal Foil Type 1	"	"	" "
" " " 2	"	"	" "
" " " 3	"	"	" "
" " " 4	"	"	" "
" " " 5	"	"	" "
" " " 6	"	"	" "
" " " 7	"	"	" "

List of Essential Oils.

1. Amyl Butyrate (Abs.)
2. Cherry Concentrate
3. Cresol Meta, C. P.
4. Essence of Strawberry
5. Ethyl lactate
6. Eugenol
7. Formic Ether (Abs.)
8. Furfural, C. P.
9. Grapefruit Concentrate
10. Grapefruit Oil
11. Guaiacol, U. S. P.
12. Lemon Concentrate
13. Lime Concentrate
14. Linalool
15. Methyl salicylate
16. Oenauthic Ether (Abs.)
17. Oil of Bergamot
18. Oil of Bitter Almonds
19. Oil of Celery
20. Oil of Cinnamon
21. Oil of Cloves
22. Oil of Lemon Concentrate
23. Oil of Nutmeg
24. Oil of Pennyroyal
25. Oil of Peppermint, U. S. P.
26. Oil of Sweet Birch

List of Essential Oils, (cont.)

27. Oil Thyme
28. Orange Concentrate
29. Pelarqonic Ether
30. Pineapple Concentrate
31. Strawberry Concentrate
32. Terpenol
33. Toly1 aldehyde.

This list of essential oils was obtained from Mr. Fitzgerald of the Frosted Foods Sales Corporation.

Section 3.

THE PENETRATION OF AIR.

The Penetration of Air.

Review of the Literature.

In a research paper by F. T. Carson many of the effects of experimental conditions on the measurement of the air permeability of paper have been outlined as follows:

The rate of flow is proportional to the differences in pressure to within a fraction of 1% except for a few of the thinnest papers.

There is very little change of rate with the time except in the case of hygrometric changes in the paper.

The quantity of air is proportional to the area exposed.

Air permeability is practically independent of the temperature.

For humidities of 65% or less there was no change in the air permeability in some papers while in others the change was as great as 15%. The direction of the change could not be predicted but seems to be entirely dependent on the particular type of paper considered.

Air permeability is inversely proportional to the thickness of the sheet.

It is not definitely known but the air may flow between the fibers of the paper or through the tubular passages of the fibers in the paper. This is probably dependent on the type of paper under consideration also. (3)

Experimental Work.

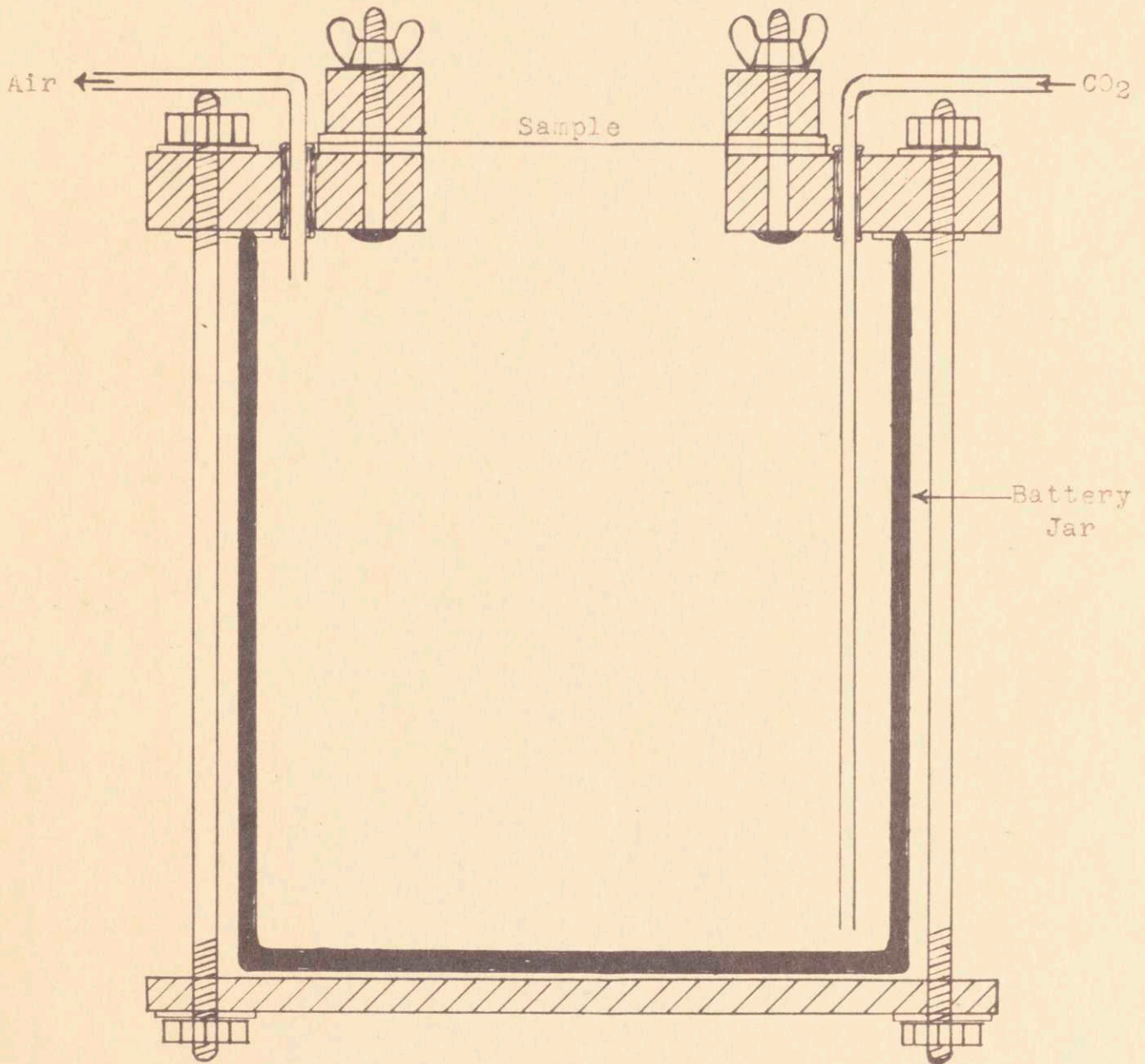
The active ingredient of air which is harmful to some food products is oxygen, so that the penetration of air itself is not nearly so important as the penetration of oxygen.

To this end some work was attempted by the use of methylene blue which would indicate the presence of oxygen. This was placed within a cell, one end of which was a sample of the wrapping material, and from which the oxygen had been removed. By the color change of the methylene blue it was hoped that varying amounts of oxygen could be detected. The color change seemed to be dependent more on the time of exposure rather than on the material through which the oxygen penetrated so a different method of attack was tried.

The principle of the new method of attack was that of measuring quantitatively, by means of a Hemple Gas analyzer, the amount of oxygen which penetrated the sample. To this end a special apparatus was constructed. Its operation can best be understood by reference to the accompanying diagram, Fig. 4. It consisted essentially of a battery jar 5 inches in diameter and 7 inches tall, the top of which was covered with a 3/4 inch board in which a round hole three inches in diameter was cut. The hole was fitted with another board, smaller than the first, also having a three inch hole in it. By means of four wing screws a sample could be clamped between the two boards. A rubber gasket was provided both above and below the sample so that an air-tight joint could be had. In addition to this there were two small holes bored through

Diagram No. 4.

Apparatus for Measuring Penetration
of Air.



the larger board, leading into the jar. These holes were fitted with rubber stoppers and glass tubes. One hole admitted a glass tube which ran to the bottom of the jar and the other a short tube leading out of the top of the jar.

In operation a sample was clamped firmly in place and then carbon dioxide was bubbled into the jar slowly to force the air out through the other opening. It was found that the jar would be very nearly free from air if the carbon dioxide were allowed to bubble in (at the rate of about three bubbles per second) for fifteen minutes. After this time the gas was shut off and a 25 cc sample withdrawn into the Hemple Gas Analyzer. The pressure was then equalized by adding a little more carbon dioxide, after which, the unit was allowed to stand four hours. At the end of this time another 25 cc sample of gas was withdrawn and analyzed as before.

In the actual analysis of the gas samples, the gas was first washed with caustic solution to remove the carbon dioxide, then it was washed with potassium pyrogallate to remove the oxygen. Since we were interested especially in the oxygen the amount of gas absorbed by the pyrogallate was taken to be the determining factor of this test. By comparison of the amounts of oxygen present, before and after the four hour exposure period, the amount of air or oxygen which penetrated the sample during the four hour period could be calculated.

At first it seemed as though the exposure period were too short because only small amounts of oxygen penetrated the sample. The exposure time was then increased to 24 hours. This gave substantially higher readings. Upon trying to check the results on a single sample it was found that very wide variations were to be expected. Unfortunately it appears that these variations are sometimes greater than the differences between the various samples so that this method in its present form can hardly be expected to give results which could be considered.

Summary and Conclusions.

The only test for the measurement of the penetration of air which was developed as a result of this work was not very satisfactory. It consisted of measuring the amount of oxygen, by means of a Hemple Gas Analyzer, which penetrated through the sample into an atmosphere of carbon dioxide. The apparatus can best be understood by reference to the diagram. The jar was filled with carbon dioxide by allowing it to bubble in slowly, thus displacing the air. After fifteen minutes a sample of the gas was taken and analyzed for oxygen. The pressure within the apparatus was then equalized with the atmospheric pressure and then the cell was allowed to stand for 24 hours after which another sample of gas was taken and again analyzed for oxygen. In trying to get check results rather wide variations were had so that this method in its present form hardly seems practical.

From the test results obtained it is impossible to

classify the types of materials studied with regard to their ability to resist the penetration of air.

Recommendations.

The principal ^{idea} of this method seems sound although the results obtained were not very good.

No attempt was made during this work to condition the samples before testing or during testing. It occurs to me that the importance of this step cannot be overstated. This should be the basis for further work and as a suggestion very different results might be obtained if the samples were conditioned at about 50% relative humidity for at least 24 hours before testing and during testing. The temperature is likely of much less importance than the humidity but it too may have some effect, thus being partially responsible for the variations now obtained.

Penetration of Air and Oxygen.

			<u>4 Hour Test Period</u> <u>of 25 cc Sample</u>	
<u>Sample</u>			<u>% Oxygen</u>	<u>% Air</u>
Cellophane	Type	1	0.8	4.0
"	"	2	1.1	5.5
"	"	3	0.9	4.0
"	"	4	1.2	6.0
Glassine	Type	1	0.9	4.5
"	"	3	1.1	5.0
Parchment	Type	4	0.7	3.5
"	"	5	0.4	2.5
"	"	6	0.8	4.0

			<u>24 Hour Test Period</u> <u>of 25 cc Sample</u>	
<u>Sample</u>			<u>% Oxygen</u>	<u>% Air</u>
Cellophane	Type	1		26.0
"	"	2		48.8
"	"	3		30.4
"	"	3		26.0
"	"	4		32.0
Parchment	Type	3		48.0
"	"	6		24.0

Not Determined.

Section 4.

THE PENETRATION OF MICROORGANISMS.

Bacteria and Molds

The Penetration of Microorganisms.

Bacteria and Molds.

Review of Literature.

W. B. Tibbetts, in his thesis "The Permeability of Cellophane to Microorganisms", stated that Plain Transparent and Moisture-proof Cellophane are impermeable to microorganisms under ordinary conditions. For his work he used seven bacteria and one mold and grew them on media such as nutrient broth, nutrient agar, malt extract agar, and peptone solution. With the liquid media he had a great deal of trouble because they caused the Cellophane to blister and fall to pieces. With solid media he had much better success so that his conclusions were drawn mostly from this phase of the work. (4)

Macy in his work found that from 10 to 12% water was necessary in the medium in order for molds to grow. He also found that below 70% moisture in the atmosphere most molds could not grow. From studies with wrapped butter he concluded that the wrapping material was undoubtedly responsible for some of the molds found in the product. (5)

Experimental Work.

First a series of tests were run to determine the numbers of microorganisms found on various samples of wrapping materials. To do this strips of the samples about $1/4 \times 1 \ 1/2$ inches were cut and placed in 10 cc of dilution water. 1 cc of this dilution water was then placed in a petri dish with nutrient agar. Counts were made on these plates at intervals of 24, 48, 72 and 120 hours.

At the same time a similar set of samples were cut and this time the strips were placed in a small desiccator, above an aqueous solution of formaldehyde for the purposes of sterilization. The samples were allowed to remain here for 24 hours after which they were plated out with nutrient agar just as in the case of the untreated samples. Counts were made on these plates at intervals of 24, 48, 96, and 120 hours. In general it was found that nearly all the samples were sterile after this 24 hour exposure to formaldehyde.

The next step was to obtain a medium which was dry enough so that it would have no harmful effect on the wrapping material samples and at the same time be moist enough to support the growth of a bacterium. In this case *Serratia marcescens* was used because of its characteristic growth. Using such an organism alone it was unnecessary to sterilize the samples, thus saving much time and work.

A gelatin medium was tried but with numerous attempts no medium was secured which was dry enough so that no harmful effects to the samples would result.

Ordinary corn meal was then tried using various percentages of water including 5, 10, 20, 30, 100, 200, and 300. The latter did support the growth of *Serratia marcescens*, but seemed to be too rough in texture to be placed in intimate contact with the sample.

Bolted corn meal was then tried using the following amounts of water: 1 part corn meal to 1, 2, 3, 4, 5, 6, and

7 parts of water. Of all these batches the one containing 6 parts of water seemed to be the best from several stand-points. First, it was wet enough to be poured when hot; second, wet enough to support a good growth of *Serratia marcescens*; and third, dry enough to have no bad effects on any of the samples from the standpoint of weakening or blistering. This medium was then used for the subsequent tests.

In the test itself, samples of the various materials, about two inches square, were cut and placed in petri dishes. These samples were all bent into the shape of an "L" by folding them about half an inch from one edge. The sample was then held in place and two small batches of the corn meal medium were poured into the plate, one on the sample in contact with the edge which had been folded up and the other in the plate in contact with the opposite side of the same edge of the sample, Fig. 5.

When the medium had cooled sufficiently the batch on the sample was inoculated with *Serratia marcescens*. All the plates were carefully examined each day for evidences of the organisms growing through the sample.

Most of the samples with which this test was tried were found to be impermeable to bacteria, if the single organism used be considered as representative of bacteria in general.

The same medium and procedure was used to determine the permeability of the samples with respect to molds. In the microbiological examination of the samples some molds



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p.39 omitted.

were found to be present. Although no special pains were taken to identify the molds found, it was observed that *Aspergillus niger* was found to be very prominent. As with the bacteria it is undesirable to sterilize the samples for test purposes. It was then desirable to use a mold which is not commonly found on the samples and for this purpose *Mucor cunninghamella* was chosen. The test system used was exactly the same as that described for bacteria except that the batch of medium placed on the sample was inoculated with a culture of the mold, *Mucor cunninghamella*. The plates were carefully examined each day for evidences of the organisms growing through the sample. In both cases an incubation temperature of 20°C was found to produce very good results.

Summary and Conclusions.

A method which was found to be satisfactory, for the determination of the permeability of food wrapping materials to microorganisms including bacteria and molds was that previously described. A two inch square sample of the material was cut and folded into the shape of an "L" by folding it about half an inch from one edge. The sample was placed, with the largest side down, in a petri dish. Then two small batches of corn meal medium were poured in, while hot, so that each was in direct contact with the short edge of the sample which projected upwards, but at no place in contact with each other. The batch of medium on the sample was then inoculated with either *Serratia marcescens*, as a representative bacterium, or *Mucor cunninghamella*, as a representative mold.

The dishes were incubated at 20°C and carefully examined each day, for about ten days, for evidences of the penetration of the organism through the sample.

The corn meal medium used was prepared by adding 60 ml. of water to 10 g. of finely bolted corn meal and, after thoroughly mixing, sterilizing the batch in an autoclave for 20 minutes at 15 pounds pressure.

Twenty-four hours exposure to formaldehyde had a definite sterilizing effect on all the members of the Cellophane and glassine types considered but did not kill all the bacteria on the metal foil samples.

All the samples except one of the glassine type were found to be impenetrable to bacteria. With the mold used all the samples except three of the glassine type and the one sample of paper type were found to be impenetrable.

<u>Sample</u>	<u>Untreated.</u>				<u>24 Hrs. in Formaldehyde Vapors.</u>			
	<u>24 Hrs.</u>	<u>48 Hrs.</u>	<u>72 Hrs.</u>	<u>120 Hrs.</u>	<u>24 Hrs.</u>	<u>48 Hrs.</u>	<u>96 Hrs.</u>	<u>120 Hrs.</u>
Cellophane Type 3	0	1	2	2	0	0	0	0
" " 4	4	7	8	9	0	0	0	0
Metal Foil 3	0	1	2	9	2	2	3	4
" " 5	1	3	3	4	2	4	5	5
Glassine 4	3	9	30	64	0	0	0	0
" 10	2	2	3	3	0	0	0	0
" 11	0	2	2	2	0	0	0	0
" 13	0	4	4	4	0	0	0	0

Bacteria Found on Various Samples and
the Sterilizing Effect of a 24 Hour
Exposure to Formaldehyde.

Penetration of Microorganisms

Bacteria and Molds.

(Observations every day for 10 days)

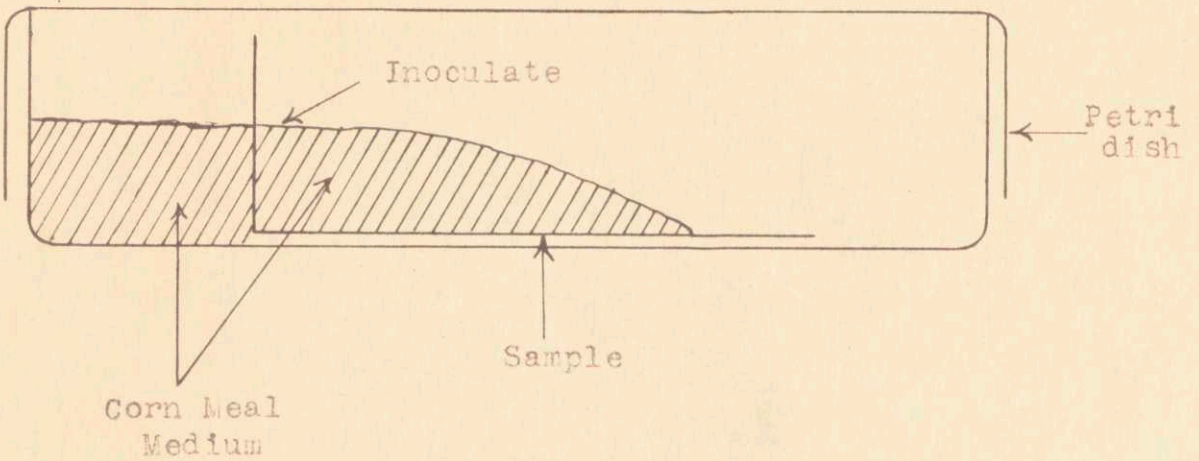
<u>Sample</u>		<u>Bacteria</u>	<u>Molds</u>
Cellophane	Type 1	No	No
"	" 2	No	No
"	" 3	No	No
"	" 4	No	No
Rubberized	1	No	No
"	2	No	No
"	3	No	No
Paper		No	Yes
Metal Foil	1	No	No
"	" 2	No	No
"	" 3	No	No
"	" 4	No	No
"	" 5	No	No
"	" 6	No	No
"	" 7	No	No
Glassine	Type 1	No	No
"	" 2	No	No
"	" 3	No	No
"	" 4	No	No
"	" 5	No	Yes
"	" 6	No	No
"	" 7	No	No
"	" 8	No	No

Penetration of Microorganisms (cont.)

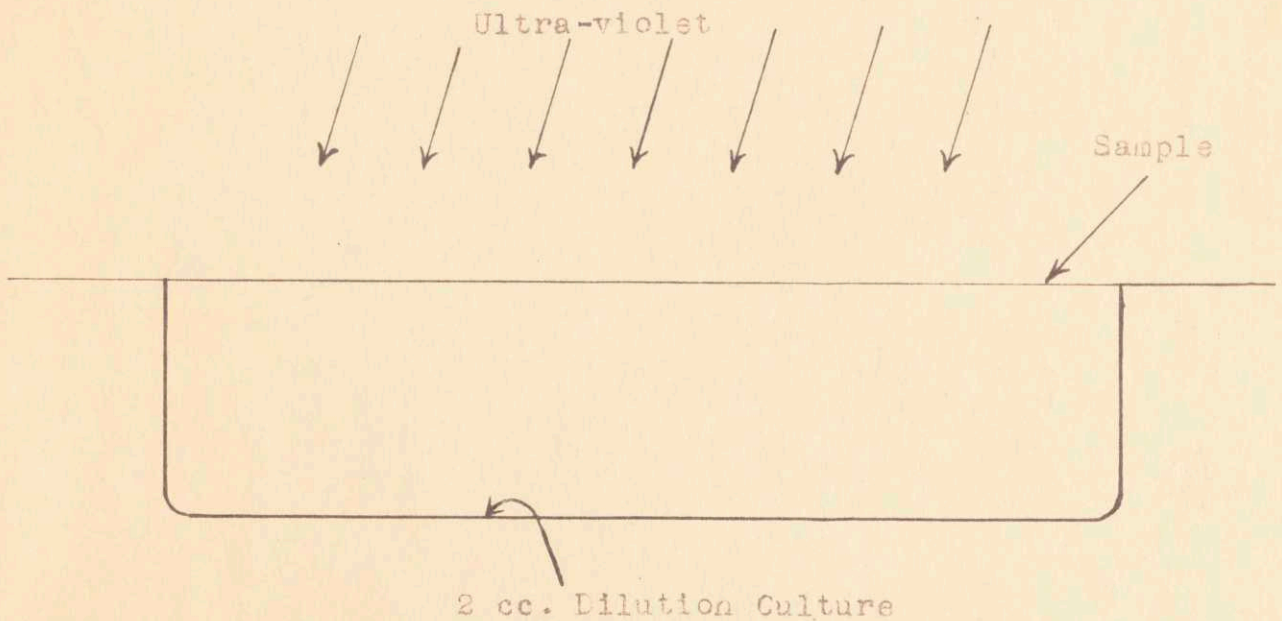
<u>Sample</u>		<u>Bacteria</u>	<u>Molds.</u>
Glassine Type	9	No	Yes
Glassine Type	10	No	No
" "	11	No	Yes
" "	12	Yes	No
" "	13	No	No
" "	14	No	No
" "	15	No	No
Parchment Type	1	No	No
" "	2	No	No
" "	3	No	No
" "	4	No	No
" "	5	No	No
" "	6	No	No

Diagram No. 5.

Apparatus for Measuring Penetration
of Microorganisms.



Apparatus for Measuring Penetration
of Ultra-violet Light.



Section 5.

THE PENETRATION OF ULTRA-VIOLET LIGHT.

The Penetration of Ultra-Violet Light.

Review of Previous Work.

Previous methods for the determination of the penetration of ultra-violet light have been chiefly chemical and biochemical methods for detecting rancidity.

The Kreis test has been rendered roughly quantitative by the adoption of color standards, but depends for its reaction on the presence of bodies of an aldehydic nature. (9) These themselves are decomposition products of the labile peroxide compounds first formed. Thus with this method the earliest stages of the rancidity cannot be investigated.

The Schibsted test depends also on the formation of aldehydes. (10) This test is essentially an adaptation of the Schiff test so that it will be slightly more sensitive. This method might well be used by determining the extent of the rancidity in samples of fats which have been wrapped in wrapping material samples and exposed, for equal periods of time, to ultra-violet light irradiations.

Experimental Work:

Ultra-violet light has an oxidizing effect on some substances and it was suggested that perhaps the color change of methylene blue solution might be used as an indication of this. To determine whether or not this were possible strips of white blotter material were saturated with the methylene blue solution and exposed to the source of ultra-violet. At first the results seemed to indicate that there was a definite color change both in the wet and dry condition. Upon further

examination it was found that the change was actually caused by the ozone generated by the lamp and not by the ultra-violet light.

The lethal effect of ultra-violet on bacteria was then tried as a criterion. At first a culture of *Serratia marcescens* was exposed to the radiations of a lamp emitting light of a wave length of 2537 Angstroms, and it was found that in five minutes most of the bacteria were killed.

In order to apply this to the determination of whether or not ultra-violet light penetrates the various samples of wrapping material a dilution culture of the bacteria was made so that two cubic centimeters of the culture contained from 25 to 50 organisms. The method used was to place two cubic centimeters of this inoculated dilution water in each petri dish, using one petri dish for each sample of material tested. Two plates were not treated in any way, two were exposed to the ultra-violet lamp with their covers removed, and the rest of the plates were exposed to the lamp using a sample of wrapping material as a cover, the regular cover having first been removed. All exposures to the lamp were five minutes in length. Melted nutrient agar was then poured into the plates, about 10 cc in each, and then they were incubated at 20°C and counts were made each day for three days. For some reason *Serratia marcescens* would not grow in the plates, as expected, including some of the control plates.

A culture of Fleischmann yeast was treated in the same way and found to respond in exactly the same way. The yeast

cells were killed in great numbers when an edge of the yeast cake was exposed to the ultra-violet lamp but in dilution in sterile water no growth was recorded in many of the control plates. If this were the only trouble it could probably be remedied by using a much more heavily seeded culture.

The main trouble seemed to be that a considerable amount of contamination resulted when the plates were uncovered and exposed to the lamp. Most of the organisms found in the air can grow very well at 20°C. In spite of all the care possible, large numbers of contaminating organisms found their way into the plates.

In some work on a thermophylic bacterium found in sugar and sugar solutions, it was found that many of the thermophylic organisms were killed by exposing a culture to the ultra-violet lamp. A culture of these was obtained and diluted so that two cubic centimeters contained from 25 to 50 organisms. This dilution culture was then used exactly in the same way as previously described except that litmus lactose agar was used as the nutrient medium and the plates were incubated at 55°C. Very reasonable results were obtained this time and no trouble was had from the contaminations from the air during exposing the plates to the lamp. The actual results are recorded at the end of this section.

Another set of tests were run one week later using the same samples and the same culture of thermophylic organisms but this time none of the organisms grew even in the control plates.

This test method seems to have possibilities of being a good one but there are several important disadvantages. First, the culture of thermophylic organisms seems to die easily, and second, it is possible that the actual wave length of light which has a lethal effect on bacteria is not the same as that which produces rancidity in fats. This latter point should be carefully investigated.

Summary and Conclusions.

The lethal effect of ultra-violet light on a culture of thermophylic bacteria, found in sugar and sugar solutions, was found to give fairly reasonable results in determining the relative penetration of ultra-violet light through samples of food wrapping materials.

The culture of a thermophylic bacterium was diluted with sterile water so that two cubic centimeters contained about 25 to 50 bacteria. Two cubic centimeters of this dilution culture were then placed in a petri dish and the dish was exposed to the radiations of an ultra-violet lamp, with a sample of the material to be tested placed over the dish, after the cover had been removed. The length of exposure to the lamp was five minutes. About 10 cc of litmus lactose agar were added and after the medium cooled sufficiently, the plate was placed, inverted in a 55°C incubator. Counts were made at intervals of 48 and 72 hours. By using several untreated controls and several controls which had been exposed to the lamp without any covering, fairly comparable conclusions can be drawn regarding the relative amount of ultra-violet which penetrated the sample.

The metal foil type of wrapping material was found to be by far the best from the standpoint of impenetrability to ultra-violet light. Next to this was the paper type and nearly all the rest seemed to have no resistance to the penetration of ultra-violet light.

Recommendations for Further Work.

Before this test method could be considered complete more work should be done on choosing a culture of thermophilic organisms which can be kept easily in culture. It might be suggested that it is well to use thermophilic organisms because at a high incubation temperature no troubles result from contaminations from the air. Most of the organisms in the air are unable to grow at temperatures as high as 55°C.

The various wave lengths of ultra-violet light which have a lethal effect on the bacteria in question, and those which cause rancidity in fats should be determined. In the event that they are not the same this test method would have to be revised considerably in order to have it indicate the penetration of ultra-violet light with respect to food materials. This point may not be especially important if a spectrum, the same as that of the sun were used. This is probably fairly constant and what we are actually interested in are the radiations which come from the sun, because this is the chief source of ultra-violet light to which food products are exposed.

Penetration of Ultra-violet Light.

<u>Sample</u>	No. of colonies on plates covered with sample and exposed 5 min.	
	<u>48 Hrs.</u>	<u>72 Hrs.</u>
Cellophane Type 1	3	4
" " 2	0	0
" " 3	4	7
" " 4	2	3
Rubberized 3	0	0
Paper	14	17
Metal Rd 2	34	35
Metal Foil 4	38	38
" " 5	36	37
Glassine Type 1	0	1
" " 2	1	1
" " 3	0	0
" " 4	0	0
" " 5	0	0
" " 7	0	0
" " 9	1	2
" " 13	0	0
" " 14	0	0
Parchment Type 2	0	0
" " 3	0	0
" " 4	0	0
" " 5	0	0
" " 6	0	0
Without sample 1	0	0
" 2	0	0
Unexposed 1	38	40
" 2	36	39

Summary of Conclusions.

The metal foil type of wrapping material was found to be the most resistant from the standpoint of penetration of moisture. Next to this type in descending order of resistance came the rubberized type, Cellophane type, parchment type, and glassine type of wrapping materials.

From the standpoint of odor and flavor penetration it was found that in general those types of wrapping materials which were resistant to the penetration of moisture were, in the same order, resistant to the penetration of odors and flavors.

No definite conclusions can be made from the standpoint of the penetration of air because in the method used the deviations between tests using the same sample were often greater than the differences between tests on the different type of materials.

The test for the penetration of microorganisms indicated that all the samples except a few of the glassine types were resistant to the penetration of *Serratia marcescens* and *Mucor cunninghamella* under the conditions of the experiment.

Using the lethal effect of an ultra-violet lamp emitting a wave length of 2537 Angstroms as a criteria for the penetration of ultra-violet light it was found that the metal foil type of wrapping material was the most resistant, and all of the other types showed practically no resistance to the penetration of ultra-violet light.

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Agencies Doing Work Relative to
the Subject.

1. American Bottlers of Carbonated Beverages,
726 Bond Building,
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2. American Management Association,
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3. American Society for Testing Materials,
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4. American Spice Trade Association,
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5. American Standards Association.
29 W. 39th Street,
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6. American Veneer Package Association,
Washington Loan and Trust Building,
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7. American Warehousemen's Association,
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8. American Waxed Paper Association,
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New York City.
9. Association Cooperage Industries of America,
2008 Railway Exchange Building,
St. Louis, Mo.
10. Bureau of Business Research,
School of Business Administration,
University of Pittsburgh,
Pittsburgh, Pa.
11. Bureau of Fisheries,
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12. Container Testing Laboratories, Inc.,
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13. The Cotton Textile Institute, Inc.,
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18. Glass Container Association of America,
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19. Institute of American Meat Packers,
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20. League for Suppression of Design Piracy,
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21. Library of Congress,
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22. Modern Packaging,
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23. National Alliance of Art and Industry, Inc.,
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25. National Cannery Association,
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34. U. S. Patent Office,
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35. Wolf and Company,
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