THE EFFECT OF HIGH MOLECULAR WEIGHT CARBOHYDRATES ON ASCORBIC ACID STABILITY IN A MODEL FOOD SYSTEM

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STEPHEN CHARLES SECREST

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ABSTRACT

THE EFFECT OF HIGH MOLECULAR WEIGHT CARBOHYDRATES ON ASCORBIC ACID STABILITY IN A MODEL FOOD SYSTEM

Product composition can affect the water activity, reactant mobility, and moisture content of a product, thus affecting ascorbic acid degradation.

This study was designed to determine the effect of fiber on ascorbic acid stability as a function of its water binding capacity. Six model systems were developed, each containing a different commercial fiber. The commercial fibers included microcrystalline cellulose (MCC), cellulose floc with 2% carboxymethyl cellulose, cellulose floc, MCC/pectin mixture (70/30), MCC/pectin mixture (85/15) and cake flour. The six model food systems were equilibrated to 0.10, 0.40, and 0.65 a_w at 20°, 30°, and 37°C, giving a total of 54 experimental conditions.

The rate of ascorbic acid degradation followed first-order kinetics under all storage conditions. Vitamin C half-life values were extended in the high water binding capacity food systems. Water activity, temperature, fiber and all interactions between the single sources of variation had statistically significant effects on ascorbic acid stability.

THE EFFECT OF HIGH MOLECULAR WEIGHT CARBOHYDRATES ON ASCORBIC ACID STABILITY IN A MODEL FOOD SYSTEM

By

Stephen Charles Secrest

THESIS

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To Jena

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INTRODUCTION

Presently, information concerning the stability of ascorbic acid as affected by water activity, moisture content, temperature and product composition is at best of limited value to the food processor. This is attributable in many instances to the use of artifically composed model systems which in no way resemble a food product. Trends, of course, can be noted when utilizing these simplistic systems, but do not always apply to an actual food product.

This study was designed to look at the above stated variables and determine what affect they would have on ascorbic acid stability in a product composed of dehydrated food components. The temperature and water activity variables were standardized to represent various storage conditions which may be found through out the United States.

Six model systems, each containing a different commercial fiber, were analyzed to measure what affect the physiochemical properties of each fiber had on the ascorbic acid degradation rate. All variables and interactions between the experimental variables were statistically analyzed to indicate which variables had the greatest and least influence on ascorbic acid stability.

REVIEW OF LITERATURE

NUTRITIONAL FORTIFICATION OF CEREALS AND FLOUR

Wheat flour, cereal based products, milk and salt have historically served as carriers for vitamins and minerals. The contributions of the vitamin enriched ready-to eat breakfast cereals and fortified flours to the dramatic decrease of beri beri, pellagra, and ariboflavinosis serve as testimony to the importance of these cereal foods as nutrient carriers.

The addition of vitamins and minerals to food products has long been viewed as a public health issue. The U. S. Food and Drug Administration, the Council on Food and Nutrition of the American Medical Association, and the Food and Nutrition Board of the National Academy of Sciences-National Research Council have therefore attempted to establish a standard nutrient fortification policy. To date, fortification policies have not been standardized because of conflicting philosophies concerning nutrient fortification. The conflicting philosophies are whether fortification should center on health needs with no concern to the composition of the original product or restoration of foods to their original nutrient levels.

Presently, nutrient fortification is carried out in a large segment of our processed foods. For most foods, fortification above the label claim is required because of

imperfect distribution of nutrients in the product, analytical error, and nutrient degradation during processing and storage. The fortification level varies with each nutrient, product, and method of application. By regulating factors such as processing temperature, method of application, product packaging material and storage temperature, maximum stability of the added nutrients can be promoted.

The food technologist is often criticized for not directing more attention to the nutitional quality of food, as affected by processing. However, the economic ease of nutrient supplementation versus changing a processing method to preserve the natural nutrients, monetarily benefits the consumer.

Presently, flour, bread, corn grits and corn meal are enriched with vitamins B_1, B_2 and niacin. Foods without standards of identity, such as ready-to-eat breakfast cereals, have been fortified with a more diverse array of micronutrients. Breakfast cereals have been fortified with as many as ten vitamins. The Food and Nutrition Board of the National Academy of Sciences - National Research Council has proposed that cereal grains (wheat, corn and rice) be fortified with vitamin A, thiamin, riboflavin, niacin, vitamin B_6, folic acid, iron, calcium, magnesium and zinc.

Extensive information concerning vitamin stability has been accumulated for selected foods. However, insufficient detail as to the effect of pH, moisture content, water activity and other product and package variables make

extrapolation to other food products difficult. Further work in this area is required.

STATE OF WATER AS IT AFFECTS MICROORGANISMS

Scott (1957) studied the effect of water on microbial growth and determined that a_w, rather than moisture content, determines the lower limit of availability of water for microbial growth. His studies indicate that most bacteria do not grow below a a_w of 0.91, most molds cease to grow below a a_w of 0.80 and xerophilic fungi are inhibited at a_w's less than 0.65 (Figure 1). Scott (1957) also concluded that environmental factors affect the specific a_w required for microbial growth. Generally, the less favorable the other environmental factors, such as nutritional adequacy, pH, oxygen pressure and temperature, the higher the minimum water activity for growth of microorganisms.

Labuza (1972) reported that, in addition to the above factors, microorganisms are affected in the following ways by the physiochemical state of the water.

1) A_w modifies the sensitivity of microorganisms to heat, light, and chemicals. In general, microorganisms are most sensitive at high a_w's (dilute solutions). Minimum sensitivity occurs in the a_w range of intermediate moisture foods (a_w 0.50 - 0.65; water content 20%–40%).

2) Minimum water activities for production of toxins are often higher than for microbial growth.

3) Microbial growth is dependent upon the method of establishing equilibrum relative humidity of a food.

Most foods show a moisture sorption hysteresis phenomenon, that is their moisture content at a given a_w is lower on the adsorption loop of their sorption isotherm than on the desorption loop. Thus, the a_w limiting microbial growth may be higher for foods reaching their equilibrium moisture content by adsorption rather than by desorption of moisture. This supports the theory that microbial growth is controlled not by a_w alone but also by total moisture content.

Some consideration has been given to the theory that differences in water content or microbial water binding may account for difference in heat resistance observed between closely related microorganisms. Tjoa (1973) noted that certain strains of Clostriduim botulinum bind more water than do other strains. The thermal resistance observed for the Clostriduim botulinum is greater in the strains that bind larger amounts of water at a given a_w.

STATE OF WATER AS IT AFFECTS ENZYMATIC REACTIONS

Acker (1962) reported that a minimum amount of water is necessary for enzymatic activity. Acker (ibid) noted that it is not the absolute water content that is the

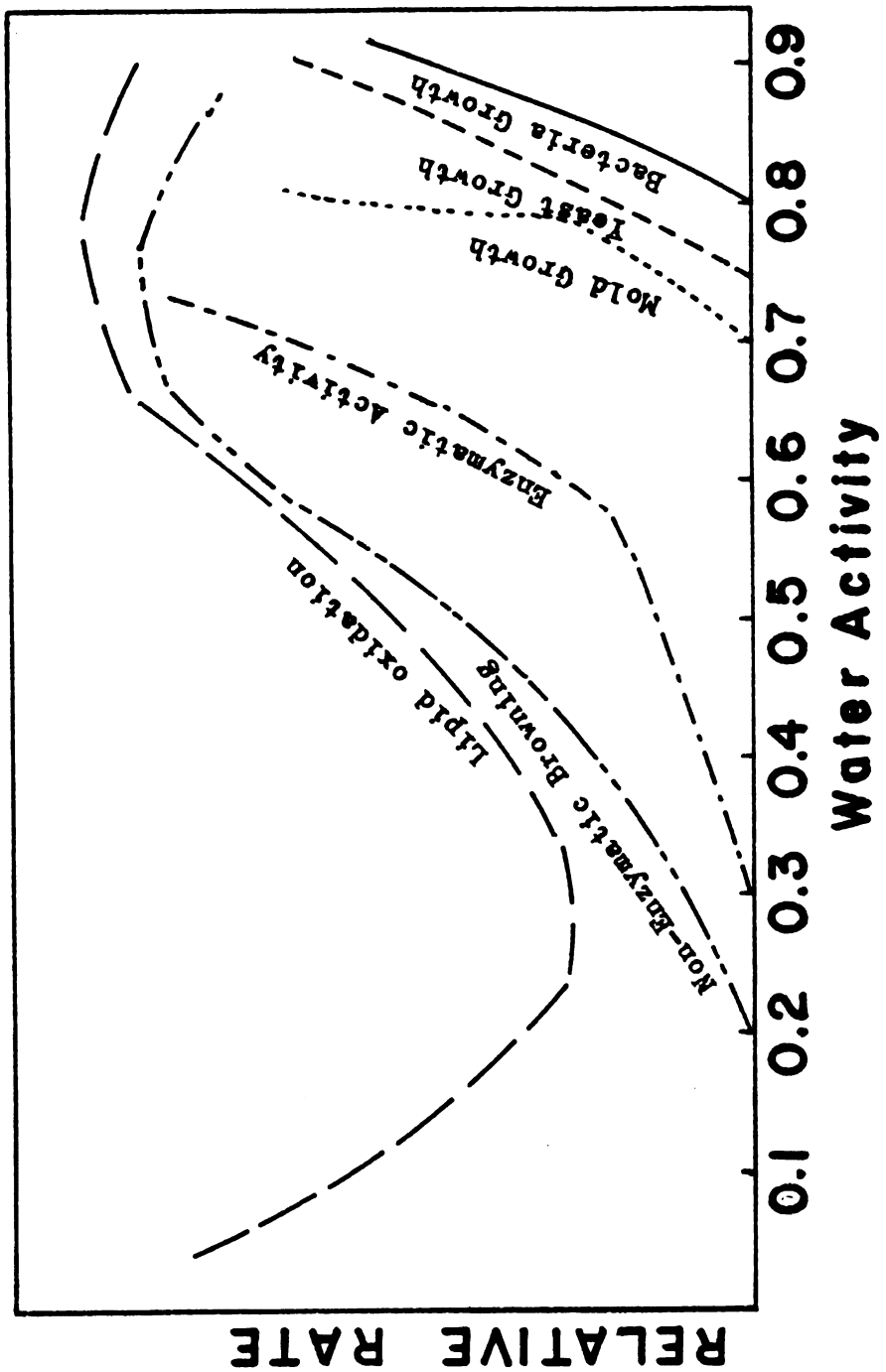

Figure 1.--Stability map of foods as a function of water activity.

controlling factor but the manner in which the water is bound. Thus a_w is more suitable than moisture content in analyzing the physio-chemical state of water and its affect on enzymatic activity. Acker (ibid) also reported that enzymatic reactions occur above the monomolecular moisture level, where water is available for reactant mobility to the active site of the enzyme. Below the monomolecular moisture level, enzymatic reactions do not normally occur because of a lack of a transport medium (Figure 1). Other factors which affect enzymatic activity as a function of a_w are:

1) The molecular weight and mobility of the substrate.
2) The heat stability of the enzyme, which is inversely related with moisture content and a_w.

STATE OF WATER AS IT AFFECTS THE CHEMICAL STABILITY OF FOOD

It is an accepted fact that the correlation between chemical reactivity of food constituents and water is best represented by the a_w of the food rather than total moisture content. An increase in availability of water usually results in an increase in chemical reaction rates, although there are some exceptions to this rule.

In chemical reactions, water can act in one or more of the following roles, as reported by Karel (1973):

1) Solvent for reactants, catalysts and inhibitors
2) Reactant (eg. hydrolysis reaction)

3) Product of reactions (eg. nonenzymatic browning condensation reactions)
4) Modifier of the catalytic or inhibitory substances (eg. inactivation of metal catlayst)

LIPID DETERIORATION REACTIONS AS AFFECTED BY WATER

Lipid oxidation, unlike most chemical and biological reactions is most rapid at low a_w's. The accelerated rate of oxidation at low a_w's is attributed to the absence of water's protective affect. Hydration of metal catalysts and hydrogen bonding to peroxides slows the oxidation reaction. The rate of oxidation decreases as the equilibrium moisture content is increased to the range of intermediate moisture foods (IMF). Salwin (1959) reported that many dehydrated products are less sensitive to lipid oxidation at a_w's above the monomolecular moisture level. The reaction rate slows with an increase in a_w ($\leq$ 0.50 a_w) where upon it will accelerate upon further addition of water.

Labuza (1970) has stated that the protective effects of water on lipid oxidation are due to:

1) Hydrogen bonding of water to hydroperoxides which are produced during the free radical reaction. This inhibits the hydroperoxides from decomposing, thus slows the rate of initiation.
2) Water lowers the catalytic activity of certain metals.

3) Water can react with trace metals to produce insoluble metal hydroxides, which removes them from the reaction phase.

At a_w's above the IMF range the rate of oxidation increases (Chou and Labuza, 1973). The acceleration of the reaction rate at high water activities (over 0.50 a_w) is due to an increase in the soluble solids content (reactants), viscosity reduction, and swelling of the polymeric matrix (Labuza, 1971). The swelling of the polymeric matrix exposes new catalyst sites thus increasing the reaction rate. At water activities approaching 0.90 the dilution of reactants and catalysts will eventually reduce the reaction rate.

The method by which foods reach a given a_w also affects the rate of lipid oxidation. Oxidation rates have been reported to be 1.5 times greater for foods equilibrated on the desorption loop of the sorption isotherm versus the adsorption loop. This difference in the rate of oxidation, when comparing the adsorption and desorption equilibrated systems, is dependent on the degree of hysteresis the food system exhibits (Chou and Labuza, 1973).

NONENZYMATIC BROWNING AS RELATED TO WATER ACTIVITY

Unlike lipid oxidation, nonenzymatic browning, which involves the reaction between the carbonyl and amino groups, increases as humidity increases to a maximum in the

a_w range of IMF and then decreases upon further dilution (Labuza, 1970).

Water is both a solvent and a product of the non-enzymatic browning reaction. At low a_w's the limiting factor is inadequate reactant mobility, however, even at a_w's corresponding to the monomolecular moisture level browning has been shown to occur (Karel and Nickerson, 1964). At very high a_w's (0.80 - 1.0) the dilution of reactants and the inhibition of browning by water condensation predominates, thus reducing the reaction rate (Labuza and Tannenbaum, 1970).

Water not only accelerates the rate of browning, but also shortens the induction period of this complex reaction (Karel and Nickerson, 1964). The effect on induction time may indicate that formation of melanoidans proceeds by a different pathway at the low a_w's or that sufficient intermediates must be formed before pigments are produced (Labuza, 1970).

STATE OF WATER AS IT AFFECTS VITAMIN STABILITY

Except for a few individual cases, extensive information relating the physiochemical state of water to vitamin stability is unavailable. However, correlations can be made between vitamin stability and the degradation of other components within the food system, which are affected by a_w. For example, the fat soluble vitamins (A,E) degrade as a function of a_w, in a manner similar to unsaturated

lipids (Figure 1).

In general, the accelerating and inhibitory effects of water on lipid oxidation reactions apply to the oxidation of fat soluble vitamins. This relationship may be an over simplification of the effects of a_w and moisture content on the lipid soluble vitamins, however, kinetic data describing the loss of fat soluble vitamins has not been completed and further conclusions can not be made at this time.

Thiamine (Mulley and Stumbo, 1975) and ascorbic acid (Lee and Labuza, 1975; Kirk and Dennison, 1977) have been studied extensively in regard to the effect of water on their rate of degradation. Their overall instability within food products can be a limiting factor in the nutritional shelf-life of foods. The degradation rates of thiamin and ascorbic acid generally follow first-order kinetics and increase with increasing a_w's. Thiamin is generally considered to be stable in most foods. Riboflavin is stable in food under ordinary conditions, however, when exposed to light this vitamin readily degrades to lumiflavin which is a strong oxidizing agent and can catalyze destruction of a number of other vitamins, particularly ascorbic acid (Tannenbaum, 1976). Ascorbic acid stability will be discussed in the next section.

Not enough is known about the other water soluble vitamins to comment on what one might expect in terms of degradation kinetics, although they are presently assumed to be more stable than ascorbic acid.

ASCORBIC ACID STABILITY AS AFFECTED BY WATER ACTIVITY AND WATER CONTENT

Water activity (a_w) is a measurement of the partial water vapor pressure of a substance divided by the vapor pressure of pure water at a constant temperature. The water absorbed by a hydrophillic food substance exists in three different states (Kupranoff, 1958).

1) The monomolecular moisture level, referring to the fraction of water which is strongly bound to individual polar groups of the food product (carboxyl or amine groups).
2) The multilayer, referring to additional layers of water molecules which are hydrogen bonded to the monomolecular layer, aldehyde and hydroxyl groups.
3) Capillary region, referring to water condensed in the interstitial pores of the food product. The lowering of water activity in this region is affected by the concentration of soluble solids (Raoults Law) and capillary action restriction.

The manner in which water is found in a dehydrated food product corresponds to three regions of the sorption isotherm of the product (Figure 2). Region A (Figure 2) corresponds to the strongly bound monomolecular moisture layer, B corresponds to the hydrogen bonded region and region C relates to the capillary condensed water. Thus a sorption isotherm would differ for each individual product

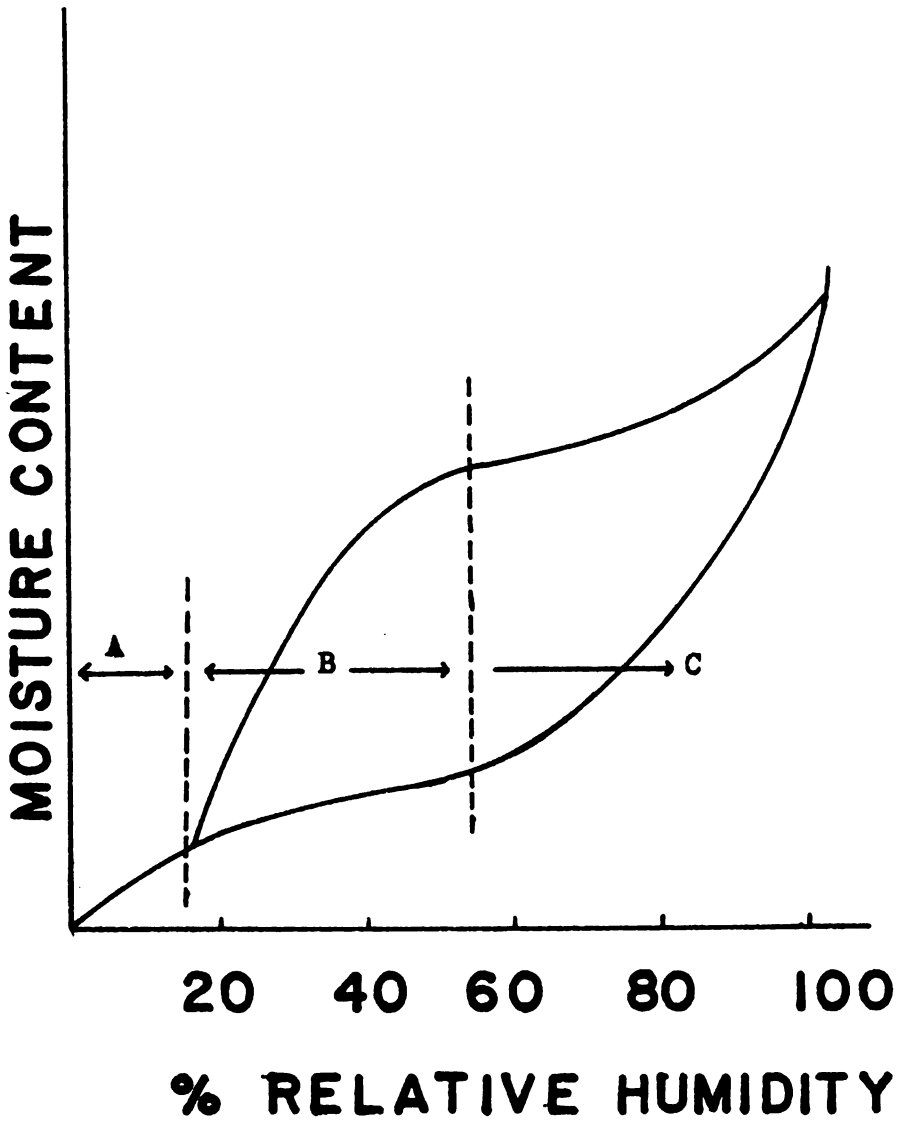

Figure 2.--Type II isotherm showing sorption hysteresis.

due to the fluctuation in water binding properties (Labuza, 1968). Because these regions overlap, we obtain the characteristic curve of the sorption isotherm.

The destruction of ascorbic acid is most rapid in the range of a_w's which include region C of the sorption isotherm (Figure 2). However, all water present, including that adsorbed in the monomolecular moisture layer appears to be available for ascorbic acid destruction (Karel and Nickerson, 1964).

At a given moisture content, the degree of water binding affects the reactant and catalyst mobility. A product having a higher proportion of ionic groups (at a given moisture content) would supposedly exhibit a lower a_w, resulting in a lower rate of ascorbic acid degradation. Likewise, at a constant a_w, the food system with a greater proportion of ionic groups would have the higher moisture content. Reactant and catalyst mobility are a function of the a_w rather than moisture content, thus at a constant a_w there is a greater loss of ascorbic acid in foods equilibrated on the desorption loop of the sorption isotherm versus the adsorption loop. This phenomena is reportedly due to the higher moisture content on the desorption loop (Lee and Labuza, 1975).

As noted earlier, an increase in a_w results in an increased degradation rate of ascorbic acid. Lee and Labuza (1975) have interpreted this increase in ascorbic acid destruction to be the result of dilution in the aqueous phase, resulting in a decreased viscosity and increased

mobility of reactants. It has also been reported that an increase in a_w or moisture content can result in swelling of the polymeric matrix, thus exposing new catalytic sites and ascorbic acid (Labuza, 1970).

The decreased viscosity resulting from increased a_w may also affect the level of dissolved oxygen in the food system. Kirk and Dennison (1977) have reported that as the a_w of a food system increases, the molar ratio of dissolved oxygen to ascorbic acid increases. This may be the primary factor responsible for the increase in the rate of ascorbic acid degradation as a function of a_w.

THE EFFECT OF PRODUCT COMPOSITION ON WATER BINDING

As stated earlier, the sorption isotherm of a food is affected by the degree and type of water binding as influenced by the food constituents. Theoretically, different food constituents could reduce or accelerate a chemical, enzymatic, or microbial reaction by altering the availability of water. The complexity of a food product results in a mixture of water binding properties, giving a continuous curve for the sorption isotherm rather than clear divisions at specific water activities. Rockland (1969) pointed out the complexity of water activity in food systems as affected by carbohydrates, proteins, lipids and minor constituents of foods.

Recent work by Leung (1976) using NMR technique, has shown that at both constant a_w's and water contents

there are dramatic differences in the tenacity with which the water is bound by different food products (Figures 3 and 4).

As shown by the data in Figures 3 and 4, pectin has a relatively strong water binding effect, whereas, water bound by cellulose was so loosely associated that it was not plotted.

Leung (1976) has indicated that as a_w increases water mobility increases (Figure 5). However, at a given a_w, there are differences in the tenacity with which the water is bound. This differs from the former theory that a_w is the ultimate measurement of water mobility.

The following list points out the degree of water binding exhibited by selected types of food components.

1) Proteins and starches adsorb more water at low a_w's than fatty materials or crystalline substances (Labuza, 1968).
2) Carboxymethyl cellulose is a better water binder than casein (Karmas and Chen, 1975).
3) Cellulose exhibits the poorest water binding capacity other than crystalline sugars (Leung, 1976).
4) Pectin binds less water than starch but holds on to the water more tenaciously (Leung, 1976).

As noted earlier the amount of water bound by a substance does not always correlate with the strength of binding. Thus product composition not only affects the amount of water bound by the system but also affects the

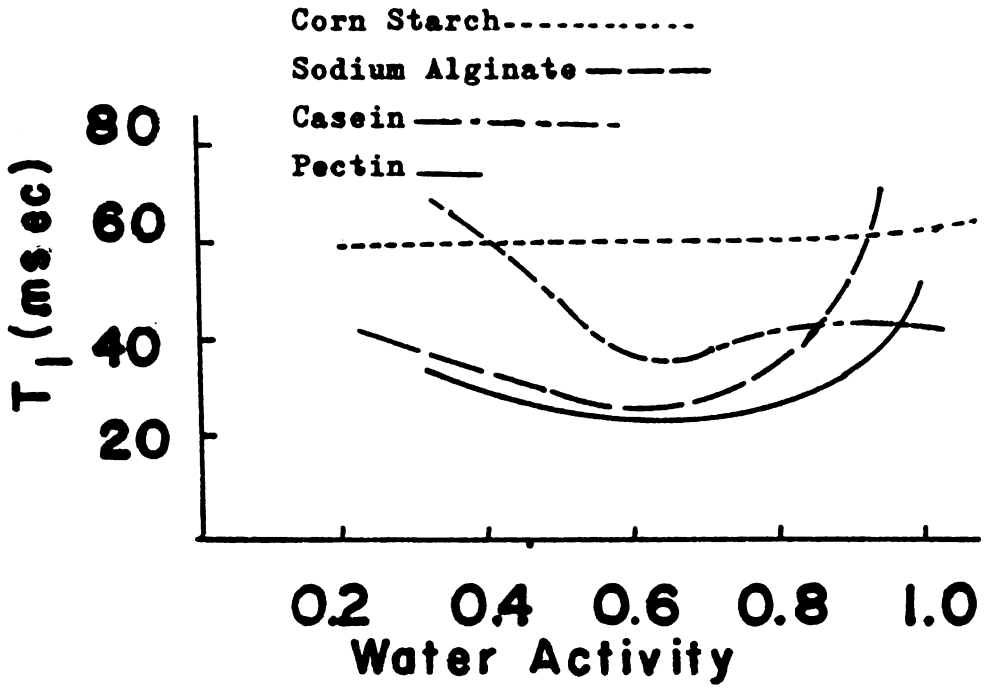

Figure 4.—Variation of spin-lattice relaxation time (T_2) with water activity for four macromolecules.

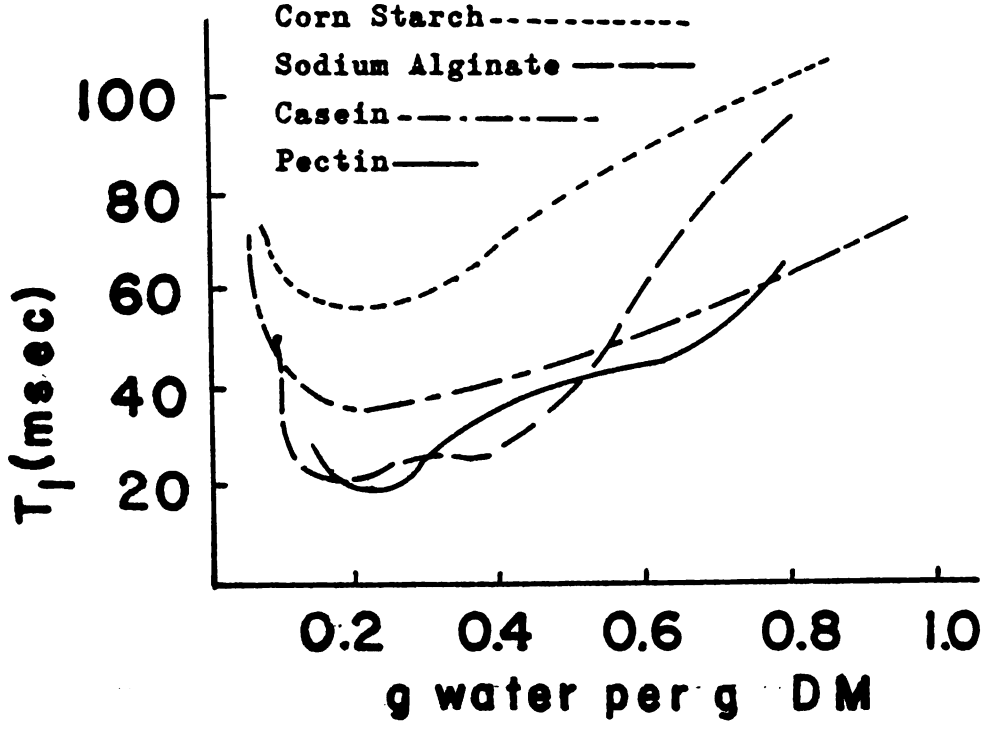

Figure 3.—Variation of spin-lattice relaxation time (T_2) with moisture content for four macromolecules.

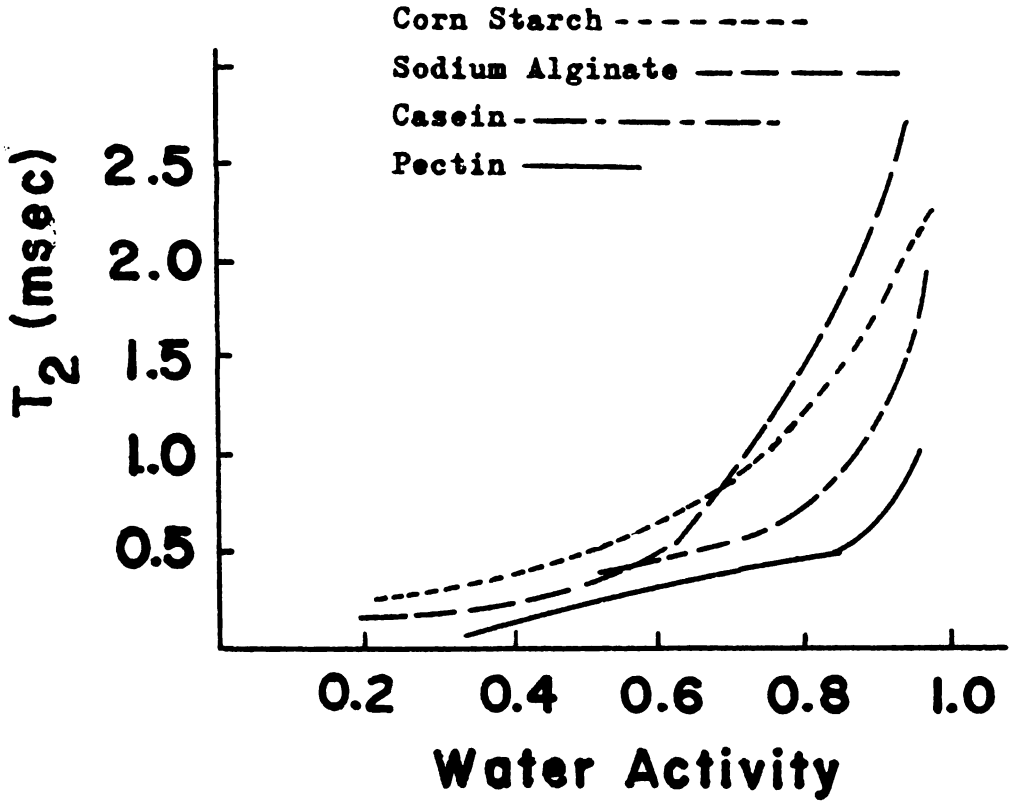

Figure 5.--Variation of spin-spin relaxation time with water activity for five macromolecules.

water mobility at both constant a_w's and moisture contents.

The effect of processing on the a_w of foods is outside the realm of this literature review, however, it should be noted that all food constituents are not affected in the same manner or degree by a single treatment. Heat, for example, has little or no effect on protein hydration, but has a dramatic effect on starch (Labuza, 1968).

FACTORS AFFECTING WATER ACTIVITY OF FOODS HAVING CAPILLARY WATER

The many differences in the chemical and physical composition of food can affect the water activity, thus influencing the degradation rate of nutrients within a food system. The capillary condensed water is most available for reactant and catalyst mobility. Therefore, factors which affect the a_w of the capillary water will have a dramatic effect on the degradation rate of ascorbic acid. The depression of a_w in the capillary condensed region is due mainly to dissolved solutes, which ideally follows Raoult's Law.

RAOULT'S LAW

$$a_w = \frac{N_w}{N_w + N_s}$$

Where a_w = water activity

N_w = number of moles of water

N_s = number of moles of solute

Deviations from Raoult's Law occur for one or more of the following reasons:

1) Not all of the water in food is capable of acting as a solvent for the solutes (eg., bound to ionic groups).
2) Not all of the solute is in solution (eg., bound to insoluble food components).
3) Interactions between solute molecules cause deviations from ideal relations.

Another factor affecting the vapor pressure at high a_w foods is depression by capillary forces. The extent to which these forces are operative at very low a_w's is not fully understood (Karel, 1973). However, one would expect definite capillary effects above 0.90 a_w if the capillaries are present in extensive numbers (Labuza and Rutman, 1968).

As earlier indicated, reactant mobility is at its maximum at the capillary condensed region. Therefore, reactant mobility could dramatically affect the stability of ascorbic acid in food systems which do not swell with water adsorption. Although reactant mobility is a rate determining factor for ascorbic acid degradation in a swelling food system, the swelling system will expose more catalytic sites upon addition of water therefore conceivably making mobility of reactants a secondary consideration (Karel, 1973).

Since mobility of reactants is affected by water binding capacity (WBC), theoretically an increase in WBC should result in a decrese in a_w. Such is not always the case. For example, in aqueous sodium caseinate mixtures there is no significant correlation between water binding and water activity (Karmas and Chen, 1975). As the concentration of sodium caseinate increases, water binding correspondingly increases, however, a_w was depressed very little. Karel (1973) concluded that this was a result of two physiochemical effects:

1) Collodial proteins do not have as much interaction with water as a completely water-soluble solute and
2) the molecular weight of proteins are too high to change the molar fraction of water and produce a significant effect on a_w (Raoult's Law).

EXPERIMENTAL PROCEDURES

MODEL SYSTEM CONSTITUENTS

Six dehydrated food model systems, differing only by the variability of commercial fiber added, were prepared according to the composition shown.

Table 1. Composition of Model Systems

Constituents	Per Cent
Flour [a]	30
Corn Oil [b]	10
NaCl	1
Sucrose	29
Commercial Fiber	30

The commercial fibers which distinguish one model system from another are listed in Table 2.

a (60% extraction) Minnill Mill, Fostoria Ohio

b Mazola Corn Oil

Table 2. Commercial Fibers Utilized In Ascorbic Acid Stability Study

Food Model System Number and Description
#1 Microcrystalline Cellulose (MCC) - FMC Corporation Low Water Retention
#2 Cellulose Floc plus 2% Carboxy Methyl Cellulose Large partical size - FMC Corporation - Prototype # 174-1 High Water Retention
#3 Cellulose Floc Large particle size - FMC Corporation - Prototype # 174-2 Low Water Retention
#4 Microcrystalline Cellulose/Pectin (70/30 ratio) FMC Corporation High Water Retention
#5 Microcrystalline Cellulose/Pectin (85/15 ratio) FMC Corporation High Water Retention
#6 Wheat Flour (60% extraction) Minnill Mills Fostoria, Ohio High Water Retention

MODEL SYSTEM PROCESSING PROCEDURE

All food model systems were processed under identical condition in order to eliminate the processing procedure as an experimental variable. The model system components (excluding fiber) were slurried to a concentration of 35% total solids in deionized water, at 60°C. The warmed slurry was homogenized at 2,500 psig on a two stage Manton-Gaulin homogenizer (2000 psig 1st stage; 500 psig 2nd stage). After

the slurry had cooled to room temperature, reduced L-ascorbic acid was added to the system to ensure homogenous distribution of the water soluble vitamin. Ascorbic acid was added at a level of 25% the NAS RDA/ 100 grams of model system (dry weight basis) or 11.25 mg/100 grams.

The slurry was then poured into freeze-drying trays (approx. 1.3 cm layer), frozen to a temperature of -50°C and freeze-dried to an absolute pressure of 5 microns at a shelf temperature of 38°C. After drying, the system was crushed and blended with the various commercial fibers using a Hobart mixer. Homogeneity of the model systems was determined by monitoring ascorbic acid concentration in various aliquots of the model system. Following mixing, the six model systems were stored in air tight containers at 4°C until samples were equilibrated to various water activities.

TEMPERATURE-RELATIVE HUMIDITY EQUILIBRATION

An Aminco-Aire unit was used to equilibrate the model systems to the desired water activities (0.10, 0.40, and 0.65). The model systems were equilibrated to their corresponding equilibrium relative humidities by placing thin slabs of the freeze-dried model system in the Aminco-Aire equilibration chamber and forcing conditioned air over the product as described by Kirk and Dennison (1977). Since all samples absorbed water during equilibration, the water activities of the sample were on the adsorption loop

of the sorption isotherm. For samples equilibrated to water activities of 0.10, a dehumidifying system in conjunction with the Aminco-Aire equilibration chamber was required to achieve the desired moisture content in the atmosphere.

A Hygrodynamics Hygrometer was used to sense the relative humidity of the air being forced through the equilibration chamber.

EXPERIMENTAL VARIABLES

Three temperatures, three water activities, and six fiber systems were used as the experimental variables in this study. For each of the six model systems there were nine storage conditions.

1) 20°C/0.10 a_w	4) 30°C/0.10 a_w	7) 37°C/0.10 a_w
2) 20°C/0.40 a_w	5) 30°C/0.40 a_w	8) 37°C/0.40 a_w
3) 20°C/0.65 a_w	6) 30°C/0.65 a_w	9) 37°C/0.65 a_w

PACKAGING OF MODEL SYSTEMS

Approximately 20 grams of equilibrated model system were placed in a 303 X 406 enameled metal can (303 can). Ample headspace was left in each container to ensure that oxygen was not a rate limiting factor in the degradation of ascorbic acid. Package variability, such as permeability to light, moisture vapor and gas transmission, was eliminated by using the metal containers. After canning, the equilibrated samples were placed in storage cubicals which

of the sorption isotherm. For samples equilibrated to water activities of 0.10, a dehumidifying system in conjunction with the Aminco-Aire equilibration chamber was required to achieve the desired moisture content in the atmosphere.

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2) $20^{\circ}C/0.40\ a_w$	5) $30^{\circ}C/0.40\ a_w$	8) $37^{\circ}C/0.40\ a_w$
3) $20^{\circ}C/0.65\ a_w$	6) $30^{\circ}C/0.65\ a_w$	9) $37^{\circ}C/0.65\ a_w$

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corresponded to the temperature at which the model system was equilibrated. Ten samples were canned for each experimental condition in order to obtain an accurate measurement of the rate of ascorbic acid destruction as a function of time.

MEASUREMENT OF ASCORBIC ACID LOSS

The model systems were removed from the storage cubicals and analyzed for ascorbic acid concentration at various time intervals depending on the rate of ascorbic acid degradation. Sample preparation included the following steps:

1) Five grams of sample were placed in a Waring blender jar (250 ml capacity) to which 100 mls of a meta-phosphoric-acetic acid solution was added.
2) Samples were blended for approximately 30 seconds.
3) Blended solutions were filtered through Whatman #42 filter paper.

Model food systems containing fibers #4 and #5 required filtration of the ascorbic acid extracts as outlined below:

Food system #4 - Filter the blended solution through Whatman #1 filter paper followed by Millipore filtration (HA 5u membrane).

Food system #5 - Filter extract through Whatman #1 filter paper followed by Whatman #42 filter paper.

Total, reduced, and dehydroascorbic acid were measured by the continuous flow o-phenylenediamine with dehydroascorbic acid resulting in the formation of a blue fluorophor, which has an absorption maximum at 350 nm and an emission maximum at 430 nm. Blank values were determined by complexing boric acid to the dehydroascorbic acid to prevent the condensation of o-phenylenediamine with dehydroascorbic acid. Total ascorbic acid was measured by oxidizing the reduced ascorbic acid to dehydroascorbic acid with 2,6 dichloroindophenol and following the same procedure described for dehydroascorbic acid.

The first ascorbic acid analysis of the model system after canning was designated as zero time when determining the degradation rate.

MOISTURE CONTENT DETERMINATION

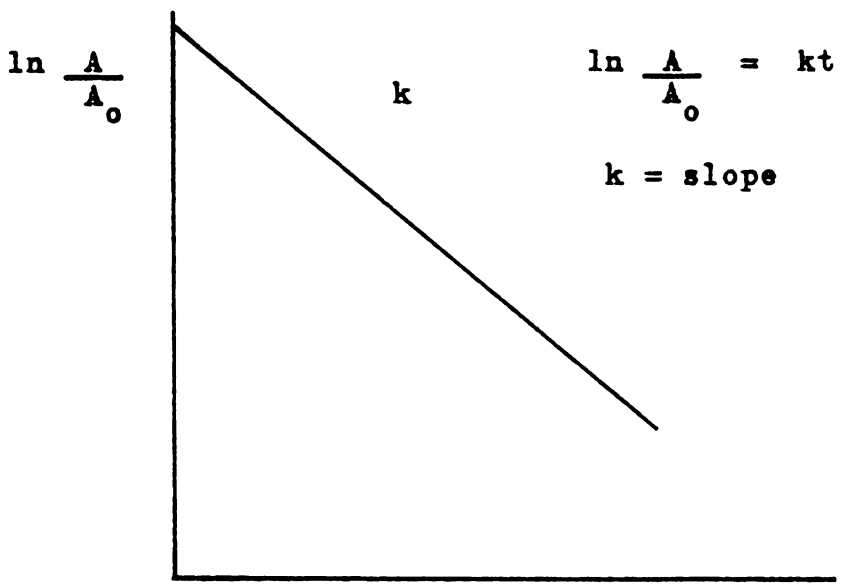
The moisture contents of the model systems were determined by placing approximately two grams of sample into a tared moisture dish and drying in vacuum at 100°C for 24 hours. A cold trap (-80°C) and the addition of dry air (bubbled through concentrated sulfuric acid) at a rate of approximately 15-20 mls/minute were used to aid in drying. Percent water was determined from the difference in weight before and after drying.

DETERMINATION OF ASCORBIC ACID HALF LIFE

The model systems were analyzed for ascorbic acid concentration as a function of time and the rate of destruction was calculated using the first order kinetics equation. The kinetic determinations were supported by data extending over at least two half lives of ascorbic acid.

The logarithim of the ratio of ascorbic acid remaining versus the initial ascorbic acid concentration was plotted against time to determine the degradation rate (k).

Figure 6.--First order kinetic plotting procedure



A = ascorbic acid concentration at various times

A_o = initial ascorbic acid concentration at 0 time

k = rate constant, $days^{-1}$

A rate constant (k) was determined for each of the 54 variable conditions. All 54 conditions followed first-order rate kinetics. The relationship between the half life of ascorbic acid ($t_{\frac{1}{2}}$) and the rate constant is as follows:

$$t_{\frac{1}{2}} = \frac{.693}{k}$$

The half life values, which are a function of the degradation rates, were used for the statistical analysis of the experimental data.

RESULTS

ASCORBIC ACID STABILITY AS A FUNCTION OF REGULATED VARIABLES

The variable conditions within the model system included six fibers, three storage temperatures, and three water activities, giving a total of 54 different conditions. As previously stated, the distinguishing factor between the six model food systems were the commercial fibers incorporated into the products (Table 2). The sources of variation in the study included fiber, temperature, a_w, fiber-temperature interaction, fiber-a_w interaction and temperature-a_w interaction.

The rate of total ascorbic acid degradation for each of the 54 conditions followed first order kinetics. The rate constants, standard deviations and correlation coeffecients are listed in Table 3. The mean correlation coefficient for the 54 rate constants was .9792. Half-life values calculated as a function of the degradation rate constants are listed in Table 4.

By studying the ascorbic acid half-lives (Table 4), one can point out specific trends due to the sources of variation.

Data in Tables 3 and 4 show specific effects of each experimental parameter on the stability of total ascorbic acid in the model food systems.

Table 3. DEGRADATION RATES FOR MODEL SYSTEMS

Condition	System #1			System #2		
	k[a]	o[b]	r[c]	k[a]	o[b]	r[c]
20°C/.10 a_w	.679	.066	.968	.586	.054	.972
20°C/.40 a_w	1.221	.051	.993	1.124	.047	.993
20°C/.65 a_w	1.518	.068	.992	1.572	.076	.991
30°C/.10 a_w	1.356	.142	.959	1.165	.122	.959
30°C/.40 a_w	2.631	.146	.988	2.403	.127	.989
30°C/.65 a_w	2.741	.075	.997	3.072	.066	.998
37°C/.10 a_w	3.320	.471	.962	2.601	.130	.995
37°C/.40 a_w	6.609	.660	.971	5.877	.557	.974
37°C/.65 a_w	7.149	.666	.979	7.452	.590	.985

a rate constant X 10^2

b standard deviation X 10^2

c correlation coefficient

Table 3. DEGRADATION RATES FOR MODEL SYSTEMS

Condition	Food System #3			Food System #4		
	k[a]	o[b]	r[c]	k[a]	o[b]	r[c]
20°C/.10 a_w	.696	.098	.945	.544	.064	.967
20°C/.40 a_w	1.572	.077	.992	.942	.052	.990
20°C/.65 a_w	1.679	.099	.986	1.339	.151	.958
30°C/.10 a_w	1.401	.117	.976	1.141	.137	.959
30°C/.40 a_w	2.721	.297	.956	1.966	.148	.981
30°C/.65 a_w	2.974	.077	.997	2.723	.192	.983
37°C/.10 a_w	2.773	.230	.987	2.259	.267	.973
37°C/.40 a_w	7.331	1.307	.929	5.188	.505	.973
37°C/.65 a_w	7.356	.572	.985	6.104	.437	.987

a rate constant X 10^2

b standard deviation X 10^2

c correlation coefficient

Table 3. DEGRADATION RATES FOR MODEL SYSTEMS

Conditions	Food System #5			Food System #6		
	k[a]	o[b]	r[c]	k[a]	o[b]	r[c]
20°C/.10 a_w	.616	.089	.943	.626	.067	.962
20°C/.40 a_w	1.037	.060	.985	1.151	.048	.992
20°C/.65 a_w	1.389	.047	.996	1.385	.069	.990
30°C/.10 a_w	1.229	.184	.948	1.148	.119	.959
30°C/.40 a_w	2.408	.082	.996	2.436	.123	.990
30°C/.65 a_w	2.915	.107	.995	2.987	.157	.990
37°C/.10 a_w	2.306	.184	.991	2.718	.361	.975
37°C/.40 a_w	6.521	.323	.994	6.252	.185	.998
37°C/.65 a_w	8.281	.623	.986	7.366	.271	.997

a rate constant X 10^2

b standard deviation X 10^2

c correlation coefficient

Table 4. ASCORBIC ACID HALF LIVES IN MODEL SYSTEMS (days)

Condition	Food System #1	Food System #2	Food System #3	Food System #4	Food System #5	Food System #6
20°C/.10 a_w	102	118	100	127	112	111
20°C/.40 a_w	57	62	45	74	67	60
20°C/.65 a_w	46	44	41	48	50	50
30°C/.10 a_w	51	60	50	61	56	60
30°C/.40 a_w	26	29	26	35	29	28
30°C/.65 a_w	25	23	23	25	24	23
37°C/.10 a_w	21	27	23	31	30	25
37°C/.40 a_w	11	12	9	13	11	11
37°C/.65 a_w	10	9	9	11	8	9

The effects of temperature and water activity on ascorbic acid stability are clearly indicated in Figures 7 and 8. The effect of the various fibers on ascorbic acid stability is not readily apparent. However, data in Table 4 indicate that fiber #4 appears to induce greater stabilizing effects on ascorbic acid than the other fibers. Model food system #4 exhibited the longest half-life values for each of the nine temperature-a_w conditions. Differences appeared to exist between the high water binding fiber systems (#2, #4, #5, #6) and the low water binding fiber systems (#1, #3). Data in Tables 3 and 4 indicate that the high water binding model food systems may have a greater stabilizing effect on ascorbic acid than the low water binding food systems (c.f., Appendix A, Fiber Group Totals). These trends require Analysis of Variance to statistically prove differences due to specific model systems and storage parameters. The statistical steps leading to the Analysis of Variance Table are found in Appendix B.

Analysis of Variance (Table 5) confirms that all sources of variation are significant. The degree at which the source of variation affects the rate of ascorbic acid degradation is indicated by the magnitude of the f-ratio. Although all single sources of variation are shown to affect the ascorbic acid degradation rate, temperature is noted to have a greater influence on ascorbic acid stability than either a_w or fiber. Likewise, a_w has a greater effect than does fiber. The interactions between all single sources of

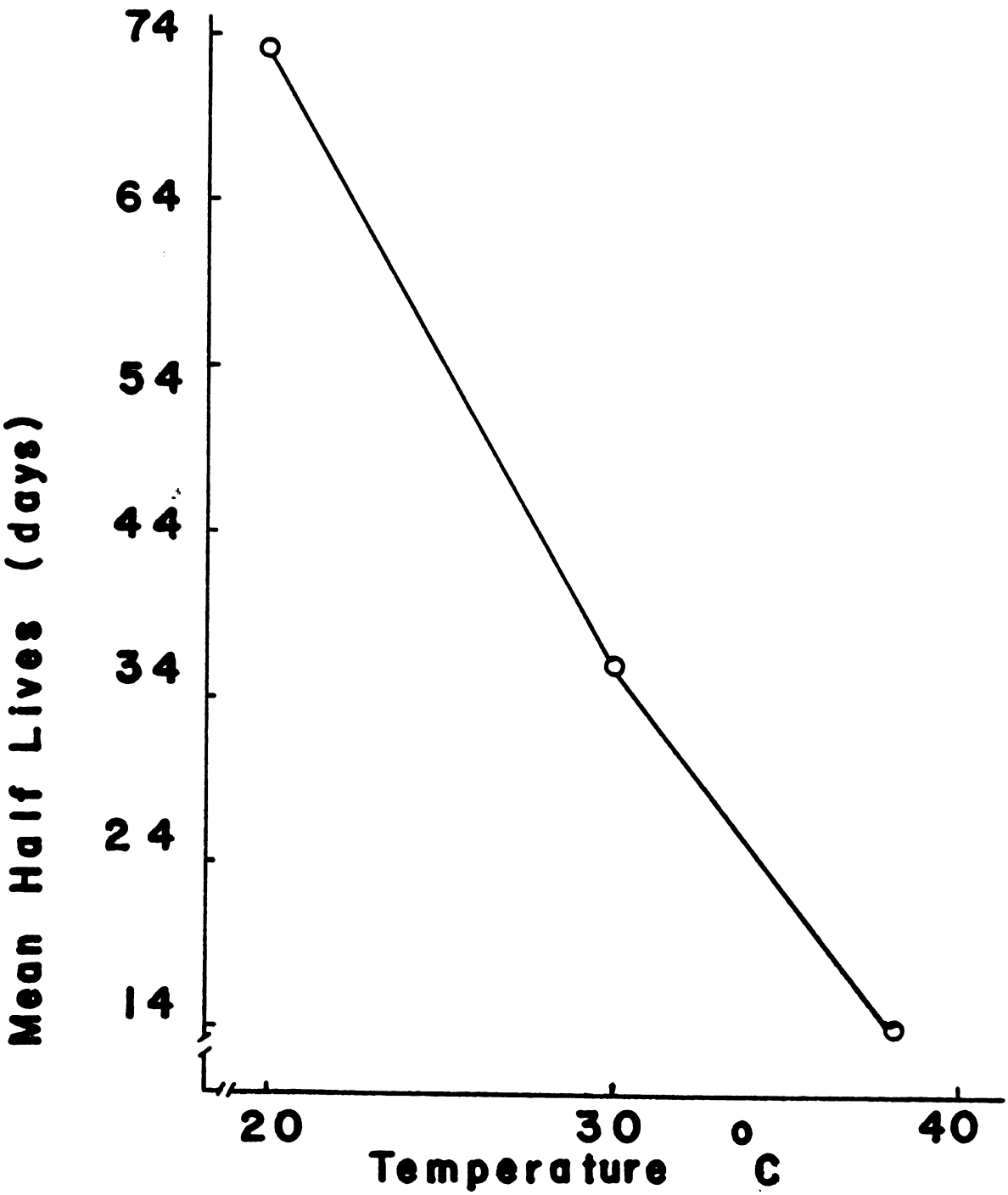


Figure 7.--Mean ascorbic acid stability as a function of temperature.

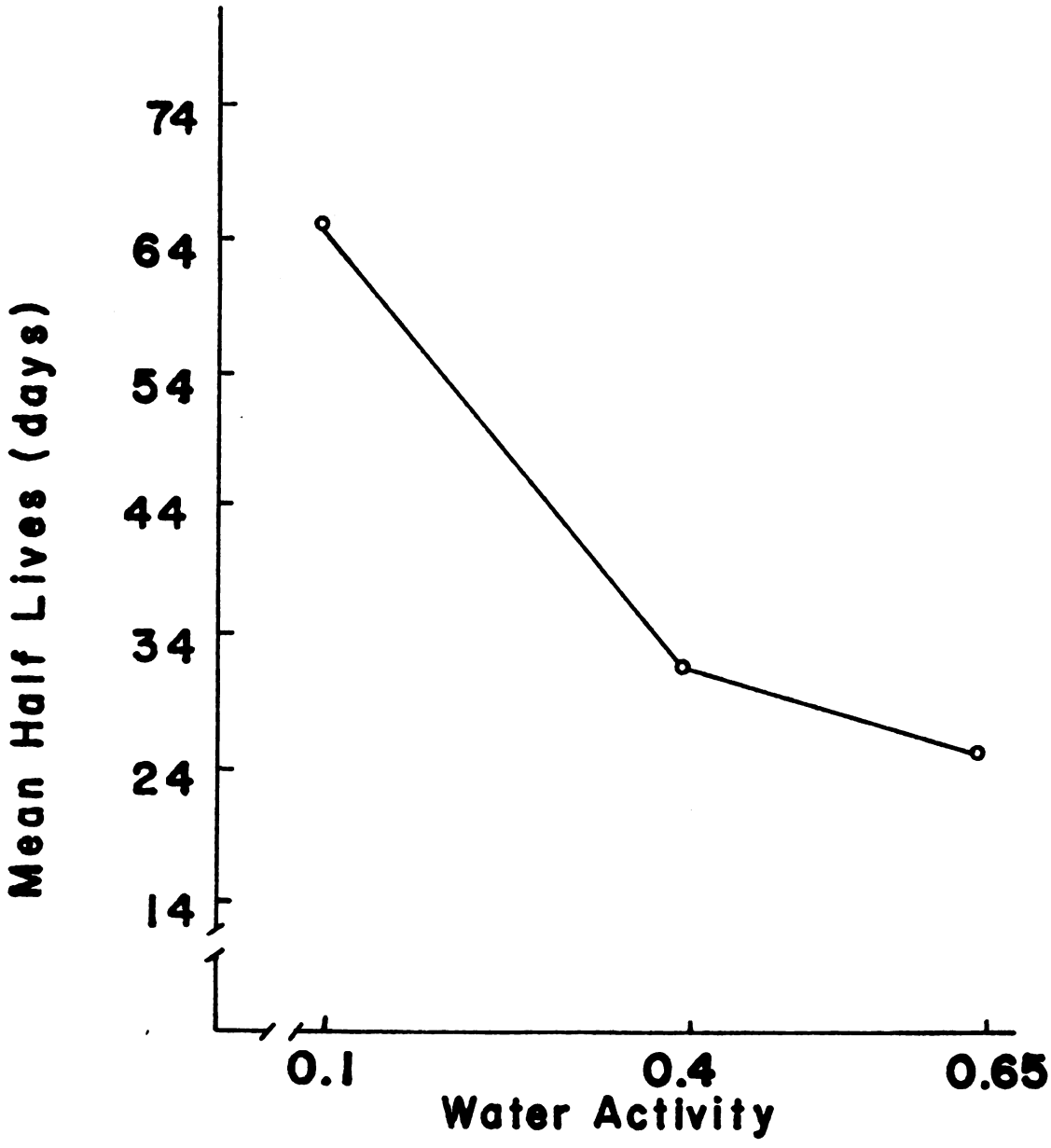


Figure 8.—Mean ascorbic acid stability as a function of water activity.

Table 5. ANALYSIS OF VARIANCE FOR ASCORBIC ACID HALF-LIFE MEAN VALUES FOR ALL MODEL FOOD SYSTEMS AS A FUNCTION OF SINGLE AND MULTIPLE EXPERIMENTAL VARIABLES

Source of Variation	d.f.	SS	MS = $\frac{SS}{d.f.}$	f ratios MS_x/MS_E	crit. Value f.05, V_1,20
A (fiber)	5	SS_A=645	129	54	2.71
B (temp)	2	SS_B=30,456	15,228	6,348	3.49
C (a_w)	2	SS_C=14,847	7,423	3,093	3.49
AB (fiber-temp)	10	SS_{AB}=273	27	11	2.35
AC (fiber-a_w)	10	SS_{AC}=245	25	10	2.35
BC (temp-a_w)	4	SS_{BC}=4,029	1,007	420	2.87
ABC (error)	20	SS_E=48	2.4		

d.f. = degrees of freedom

MS = means square

variation have been shown to have a significant effect on the ascorbic acid stability. As indicated by the magnitude of the f-ratios, the Temperature-Water Activity Interaction has a greater influence on ascorbic acid stability than do the Temperature-Fiber or Fiber-Water Activity Interactions.

As previously stated, the different fibers in the model systems appear to influence the ascorbic acid degradation rate (Table 4). Analysis of Variance confirmed that these differences are statistically significant, however, it does not indicate which fibers or why these fibers extend the nutrient half life. The Fiber Group Totals (Appendix A) indicate that the fiber in food system #4 has the greatest stabilizing effect on total ascorbic acid, followed by #5, #2, #6, #1 and #3. Food systems #2, #4, #5 and #6 all contain high water binding capacity fibers which appear to induce a more favorable environment for ascorbic acid than do the low water binding capacity fiber systems.

It is of interest to note the difference between the ascorbic acid half-life values for food systems #2 and #3 (Fiber Group Totals, Appendix A). This difference could only be attributable to the 0.6% carboxymethyl cellulose which was present in model food system #2.

In order to determine why some fibers stabilized ascorbic acid to a greater extent than others, a Bonferroni t test (Miller, 1966) was utilized.

The questions that arise include the following:

1) Is there a statistically significant difference (total ascorbic acid half-life values) among the high WBC fibers (#2, #4, #5 and #6) and the low WBC fibers (#1 and #3)?
2) Is there a significant difference (total ascorbic acid half-life values) in the low pH model food systems (#4 and #5) versus the high pH model food systems (#1, #2, #3, and #6)?
3) Is there a statistically significant difference in total ascorbic acid half-life values between the two low water binding food systems?
4) Is there a significant difference in total ascorbic acid half-life values between the two low pH food systems?

Bonferroni t test (Miller 1966)

Question #1

$$t_b = \frac{2(\bar{y}_1 + \bar{y}_3) - (\bar{y}_2 + \bar{y}_4 + \bar{y}_5 + \bar{y}_6)}{\sqrt{\Sigma C^2 (MS_E) / r}}$$

C = questions

r = repetition

MS_E = means square error

$\bar{y}_n$ = mean half life of ascorbic acid in corresponding fiber system (nine conditions)

$$t_b = \frac{2(38.7 + 36.2) - (42.7 + 47.2 + 43.0 + 41.9)}{\sqrt{\frac{12\ (2.4)}{9}}}$$

$t_b = -13.97$ vs. $\pm t_{b\ 0.025}$ 2.74

(corresponds to 95% confidence level)

Because 13.97 is greater than 2.74, the high water binding food constituents significantly enhance the enhance the environment for ascorbic acid stability.

Question #2 Low versus High pH Food Systems

$t_b = 11.68$ vs. $\pm t_{b\ 0.025}$ 2.74

Because 11.68 is greater than 2.74, polygalacturonic acid did significantly stabilize the ascorbic acid in the model food system.

Question #3 Food System #1 versus #3

$t_b = 1.17$ vs. $\pm t_{b\ 0.025}$ 2.74

In this case, 1.17 is less than 2.74, indicating no significant differences between the two low water binding food systems.

Question #4 Food System #4 versus #5

$t_b = 2.35$ vs. $\pm t_{b\ 0.025}$ 2.74

No significant difference could be determined between the two pectin based food systems.

These data confirm that by increasing the water binding capacity (WBC) of a product at a given water activity, the half life of ascorbic acid can be increased. Likewise, the addition of a food component which lowers the pH of the system may result in an extended half life for ascorbic acid. Using these two parameters it is possible to speculate that the pectin based model systems, which have lower pH's and higher WBC's, would have a greater stabilizing effect than the other fiber model systems. Experimental data confirms that the pectin based fiber systems stabilize the ascorbic acid more effectively than the remaining fiber systems.

In contrast, fiber systems which had a high pH and low WBC properties were shown to reduce ascorbic acid stability (Fiber Group Totals, Appendix A).

ASCORBIC ACID STABILITY AS AFFECTED BY INTERACTIONS BETWEEN SINGLE SOURCES OF VARIATION

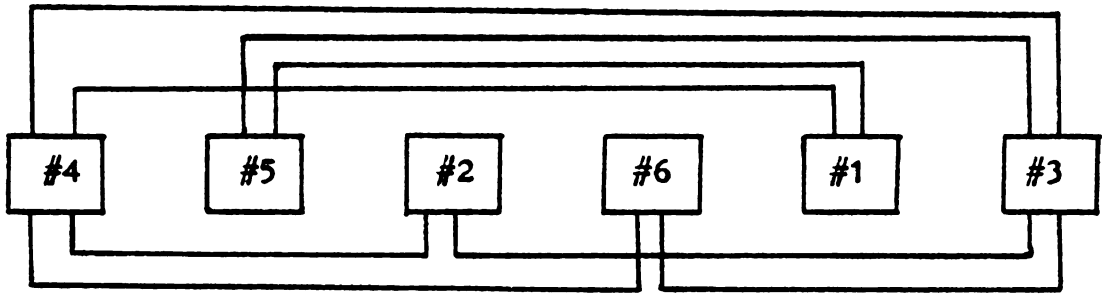
Analysis of Variance data in Table 5 confirms that the fiber-temperature interaction has a significant effect on ascorbic acid stability. However, which of these parameters, temperature or fiber, has the greater influence on this interaction? The Scheffe test (Miller, 1966) was used to determine whether the fiber has a significant affect at a constant temperature and whether temperature has a significant effect at a constant fiber (Fiber-Temperature Interactions, Appendix A).

Fiber-Temperature Interaction

Constant Temperature 20°C

As can be seen by the data (Appendix C), the effect of fiber on the fiber-temperature interaction at 20°C indicates that temperature fluctuations had a greater effect on the ascorbic acid degradation rate than did fiber variability. Data in Figure 9 indicate the food systems which are significantly different from one another (fiber-temperature interaction) as a result of fiber variability.

Figure 9.--Effect of fiber on the fiber-temperature interaction at 20°C.



Most Stable Least Stable
Fiber-Temperature Interaction
As Affected by Fiber At 20°C

Fiber-Temperature Interactions

Constant Temperature 30°C and 37°C

The Scheffe test (Miller, 1966) indicates that none of the rate constants at 30°C or 37°C were significantly different as a function of fiber variability (Appendix C).

Fiber-Temperature Interaction

Constant Fiber

With fiber as a constant and temperature as the dependent variable, it is possible to determine the degree of influence exerted by temperature on the fiber-temperature interaction. The Scheffe procedure (Miller, 1966) confirms that all of the fiber-temperature interactions are significantly different from one another as a function of temperature (Appendix C). Therefore, the effect of a fiber-temperature interaction on ascorbic acid is due more to temperature variation than fiber variability. The effect of fiber on ascorbic acid stability was shown to be more pronounced at the lower temperatures (Figure 10).

Fiber-Water Activity Interaction

Constant Water Activity 0.10 a_w

The fiber-a_w interaction is also shown to have a significant affect on ascorbic acid stability. Utilizing the Scheffe procedure (Miller, 1966), with water activity as a constant, the effect of fiber variability on the fiber-a_w interaction was determined (Appendix C). Food systems which are significantly different at their fiber-water activity interaction point, as a function of fiber variability, are indicated by attached lines.

Figure 10.--Mean ascorbic acid half lives at constant fiber and plotted as a function of temperature.

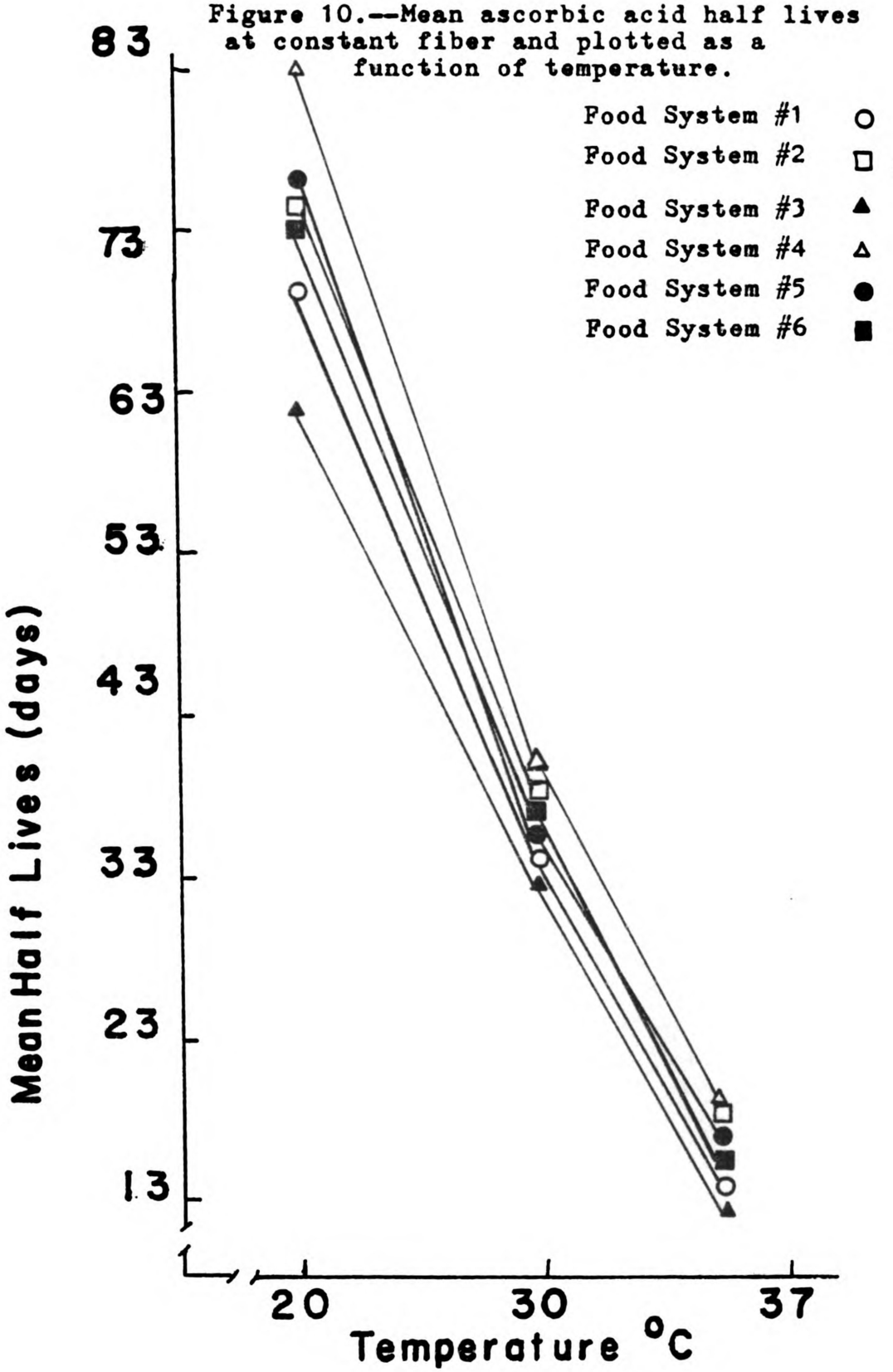
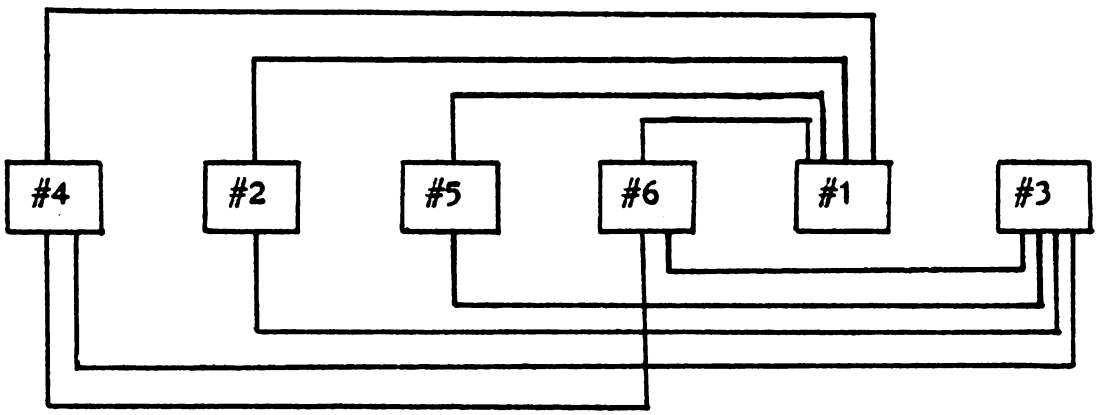


Figure 11.--Significant differences in fiber-water activity interactions as a function of fiber at a constant a_w of 0.10.



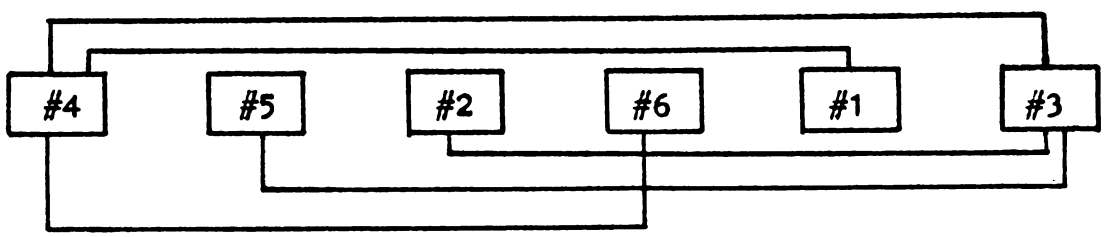
Most Stable — Least Stable
Fiber-Water Activity Interaction (0.10 a_w)

The high water binding capacity food systems (#2, #4, #5 and #6) are significantly more protective toward ascorbic acid than the low water binding capacity food systems.

Fiber-Water Activity Interaction

Constant Water Activity 0.40 a_w

Figure 13.--Significant differences in fiber-water activity interactions as a function of fiber at a constant a_w of 0.40.



Most Stable — Least Stable
Fiber-Water Activity Interaction (0.40 a_w)

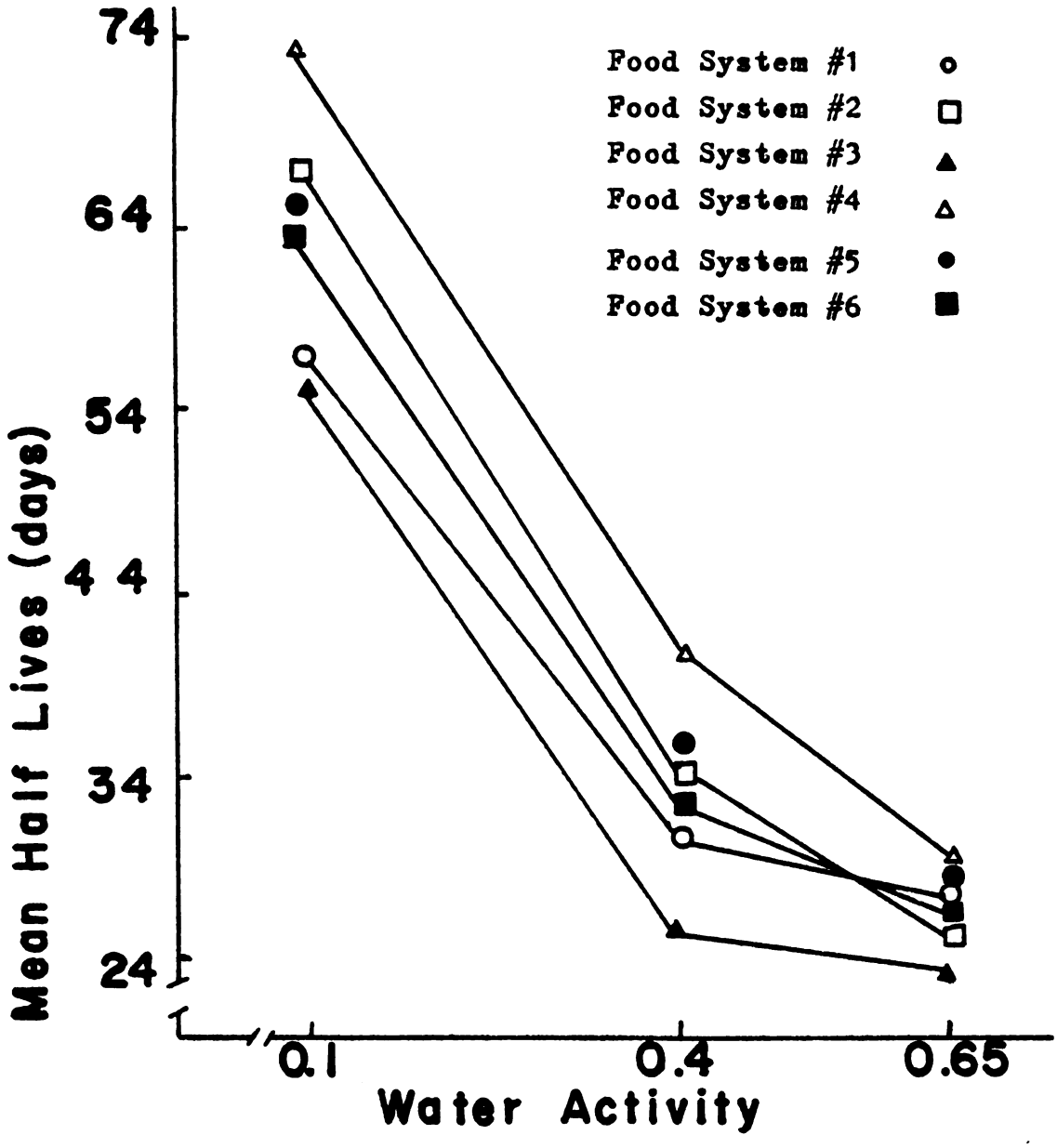


Figure 12.—Effect of the Fiber-Water Activity Interaction on ascorbic acid stability.

As noted from the data in Figures 11 and 13, there are fewer food systems at 0.40 a_w which exhibit significantly different rate constants for total ascorbic acid destruction, than at 0.10 a_w. At 0.65 a_w the stability of ascorbic acid was not significantly different in the model food systems as a function of fiber. The high WBC fiber systems did, however, correspond to longer half-life values than did the low WBC fiber systems.

FIBER-WATER ACTIVITY INTERACTIONS AS A FUNCTION OF WATER ACTIVITY

With water activity as the experimental variable, statistical analysis using the Scheffe procedure (Miller, 1966) indicates the effect of water activity on the fiber-water activity interaction (Appendix A). As shown by these data (Appendix C), all values are greater than the minimum significant difference and therefore, all points (at a constant fiber) are significantly different from one another as a function of a_w.

TEMPERATURE-WATER ACTIVITY INTERACTION

The temperature-water activity interaction did exhibit a trend in stabilizing ascorbic acid in the model food systems (Figure 11; Appendix A). Analysis of Variance of the rate constants for ascorbic acid degradation showed statistically significant interactions between temperature and

a_w, although not for all points. Using the Scheffe procedure (Miller, 1966) the effect of a_w and temperature were separately analyzed to determine their effect on the temperature-a_w interaction (Appendix C). As can be seen from the data in Appendix C, all points at a given temperature were significantly different with the exception of 0.40 and 0.65 a_w at 37°C.

AFFECT OF MOISTURE CONTENT ON ASCORBIC ACID STABILITY

Moisture contents of the model system were measured at each of the nine water activity-temperature conditions.

Table 6.--Moisture contents of the model food systems (%)

Food System	20°C/.10 a_w	20°C/.40 a_w	20°C/.65 a_w	30°C/.10 a_w
#1	2.36	3.54	5.46	2.56
#2	2.16	3.93	5.60	2.71
#3	2.30	3.74	5.55	2.98
#4	2.65	3.87	6.06	2.99
#5	2.32	3.74	5.77	2.99
#6	3.51	4.99	6.75	3.51

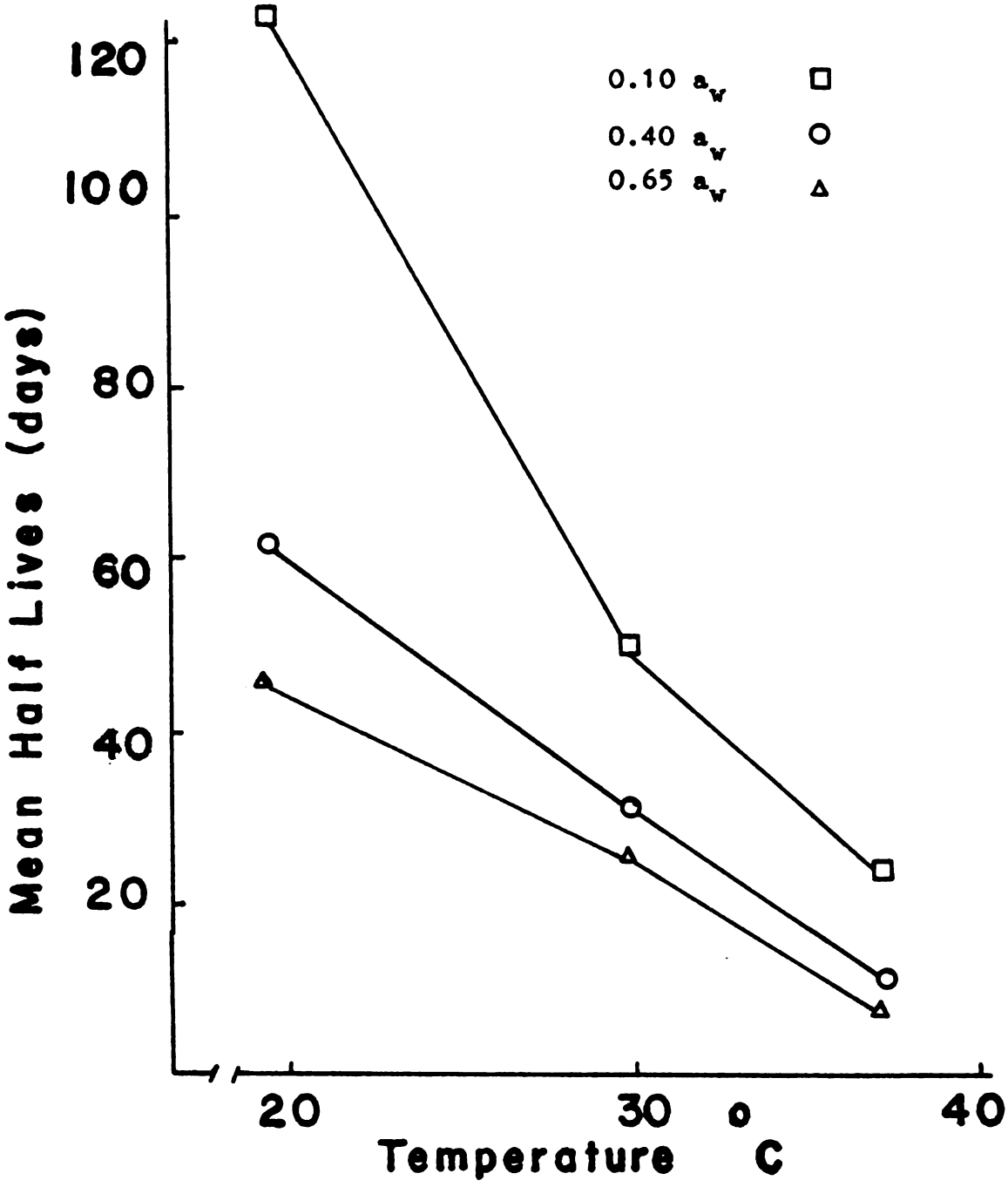


Figure 14.--Effect of temperature-water activity interaction on ascorbic acid stability.

Table 6 continued

Food System	30°C/.40 a_w	30°C/.65 a_w	37°C/.10 a_w	37°C/.40 a_w
#1	4.37	5.36	2.25	3.21
#2	4.79	5.40	2.14	3.15
#3	4.60	5.50	2.08	3.23
#4	5.17	5.98	2.40	3.68
#5	4.52	5.63	2.08	3.60
#6	5.87	7.19	2.97	5.61

Food System	37°C/.65 a_w	Mean of the nine conditions
#1	3.84	3.66
#2	3.82	3.75
#3	3.68	3.74
#4	4.43	4.14
#5	4.55	3.91
#6	5.46	5.10

Moisture contents of the model systems varied with the type of fiber incorporated. The four highest moisture contents correspond to the four high water binding capacity fibers. As stated earlier, the high WBC fibers extended the half life of ascorbic acid in the dehydrated model food systems.

The dramatic difference in water content might be expected to effect the ascorbic acid in the dehydrated model food systems. However, in food system #6 the higher moisture content did not adversely affect the stability of ascorbic acid.

DISCUSSION

The rate of degradation for total ascorbic acid, followed first-order kinetics in all 54 experimental conditions. An Analysis of Variance indicated that all single sources of variation (temperature, fiber and a_w) significantly influenced the ascorbic acid degradation rate. The degree of influence each parameter induced is a function of the range of that particular parameter. For example, if a_w had ranged from 0.00 to 1.00, it rather than temperature could have exhibited the greater influence on ascorbic acid degradation.

Of all single sources of variation, fiber variability was shown to have the least effect on ascorbic acid stability. However, as is noted by the Analysis of Variance, fiber variability does have a statistically significant effect on the rate of ascorbic acid degradation, at a constant a_w and storage temperature (Table 4). The effect of fiber on the stability of ascorbic acid appears to be related to the water binding capacity (WBC) exhibited by each fiber system. The high WBC fiber systems contained higher moisture contents than did the low WBC systems (Table 6). Not only did the higher WBC systems bind more water at a given a_w, but also significantly extended the half-life values of ascorbic acid over the low WBC food systems (Fiber Group Total, Appendix A; Bonferroni t test). This phenomenon may be due to a decreased mobility of

catalysts and ascorbic acid. This theory is supported by Leung's work (1976) which indicates that at a constant a_w, water mobility varies as a function of the food components. His work indicated that pectin drastically reduced water mobility as compared to other components, even at higher moisture contents. Thus, the same effect could apply to the pectin containing food systems in this study. Even at higher moisture contents, the pectin based food systems were most effective in extending the ascorbic acid half-lives (Tables 4 and 6).

A significant difference in the rate of ascorbic acid destruction was found to exist between the low pH model food systems (#4 and #5) and the high pH model food systems (#1, #2, #3 and #6). Whether this difference was due to the lowering of pH by the polygalacuronic acid units, an increase in WBC or metal chelation by the organic acid can not be determined from this study. No statistically significant differences in ascorbic acid degradation rates were found to exist between the two low pH food systems, nor between the two low WBC food systems (Bonferroni t Test).

The effect of fiber variability on the rate of ascorbic acid destruction was much more pronounced at low a_w's than at high a_w's. Likewise, the effect of fiber on ascorbic acid stability was more profound at low temperatures than at high temperatures. The method by which fiber affected the ascorbic acid half life values may not have changed as a function of variable temperature and a_w, but

became more noted as the influence of the dominating variables decreased.

This theory is supported by the fact that at high a_w's, differences in mobility as affected by fiber variability would be less of a differentiating factor, due to the presence of capillary water. When capillary condensed water exist, differences in water mobility as affected by product composition would be diminished. At these higher a_w's, all catalysts and reactants would be mobile, thus equilizing the reaction rates. The ability of water at low a_w's to exert vapor pressure but inability to act as a solvent for reactants and catalysts, may explain the larger fluctuations in half life values at the low a_w, as a function of WBC.

It is of interest to note that fiber variability at 0.40 a_w, is more influencial in determining the ascorbic acid degradation rate, than at 0.10 or 0.65 a_w, as indicated by the percent variation in ascorbic acid half-life values. However, at 0.10 the half-life variation in total days was greater. At 0.40 a_w all ascorbic acid and reaction catalysts may be concentrated in solution, which could influence chelation of metal catalysts by organic acids (pectin fiber). Chelation of metal ions would promote ascorbic acid stability, whereas solubilization in the concentrated form would increase the degradation of ascorbic acid. These opposing effects could be the cause for the more diverse ascorbic acid half-life values

at 0.40 a_w as a function of fiber.

At 0.65 a_w, it can be postulated that only a small amount of additional catalyst and/or reactant molecules are being drawn into solution and a dilution of reactants would occur. Thus a leveling off of the degradation rate would be anticapated. The fact that ascorbic acid degradation was less in the high WBC fiber systems (higher moisture content at a given a_w), may further indicate that the dilution of ascorbic acid and reactants as a function of fiber variability is important in the determination of the ascorbic acid degradation rate.

From these experimental data, it could be inferred that the high WBC fiber systems may produce a much more stable environment for ascorbic acid at a constant water content than a low WBC fiber system. Differences in ascorbic acid half life values, as affected by fiber variability, ranged from 27 days at favorable storage conditions (low temperature and low a_w) to only 2 days at the unfavorable storage condition (high temperature and high a_w). Whether these differences in ascorbic acid stability, as influenced by fiber variability, are sufficient for industrial application is questionable. WBC of a product, as it effects moisture content and nutrient stability, should not be underscored as a tool for the food industry, because significant economic gains can be made by selling a product with a higher moisture content which meets a_w specification.

Lee and Labuza (1975) proposed an increased mobility of reactants and catalysts was the major reason for increased ascorbic acid degradation, as a function of a_w. Using NMR, it was shown that an increase in water content resulted in decreased viscosity, which increased the mobility of water in the system. Data in Tables 4 and 6 supports the theory that viscosity plays a major role in regulating the ascorbic acid degradation rate, since even at higher moisture contents, the high WBC fibers extended the ascorbic acid half life.

As stated in the literature review, lipid oxidation is more prevalent at low a_w's than at the higher a_w's. The free radical formation which occurs during lipid oxidation, could possibly have accelerated the ascorbic acid degradation rate at the lower a_w's. However, as is evident from the data in Table 4, if lipid oxidation affected the rate of ascorbic acid destruction, it did not appear to be the predominate factor.

As expected, an increase in a_w within a single food system held at a constant temperature resulted in an increased moisture content (Table 6). Likewise, at a constant a_w and fiber, an increase in temperature resulted in a decreased moisture content (Table 6). The major differences in total moisture content at a given a_w did not noticeably effect the ascorbic acid degradation rate (Fiber Group Totals, Appendix A), indicating that a_w is a more accurate method of measuring the effect of water on the

vitamin, than is total moisture content. However, as noted by the deviations of ascorbic acid half life values, a_w is only one parameter in determining the nutrient half life in a product. Water binding capacity of the product is also of importance in predicting the ascorbic acid half life value.

A_w in the range measured (0.10-0.65 a_w) had a greater influence on the degradation rate of ascorbic acid than did fiber variability (Analysis of Variance), but less of an effect than temperature. An increase in a_w at a constant temperature and fiber resulted in a decreased ascorbic acid half life. This coincides with studies by Vojnovich (1970), Karel and Nickerson (1964), Jensen (1967) and Kirk (1977) which indicated that an increase in moisture or a_w resulted in an increased ascorbic acid degradation rate.

Lee and Labuza (1975) have noted that the mechanism involved in ascorbic acid degradation, did not change in the ascorbic acid degradation rate. Lee and Labuza's data are supported by this study which found no significant differences in the E_a for ascorbic acid destruction in the 0.10 - 0.65 a_w range (Appendix B).

Lee and Labuza (1975) speculated that with an increase in a_w above a certain level, a dilution of the reactants may be occurring. This was suspected since above a critical moisture level, the rate increase was very small and almost constant. These findings are supported by data from this study (Table 4), which indicates that above 0.40 a_w only a

slight increase in degradation rates were observed.

An increase in storage temperature at a constant a_w and fiber source was shown to have the greatest effect on the stability of ascorbic acid in the model food systems. Characteristic of most biological systems, a 10°C increase in storage temperature represented an approximate doubling of the rate of ascorbic acid destruction regardless of the fiber added to the model system (Table 4). As noted by the constant E_a, (Appendix B) the mechanism by which temperature accelerated the degradation rate did not appear to change by the increase on storage temperature.

INTERACTIONS BETWEEN SINGLE SOURCES OF VARIATION

The Analysis of Variance also points out that all interactions between the single sources of variation had a significant effect on the ascorbic acid degradation rate. The temperature-a_w interaction had a greater influence on ascorbic acid degradation than did the other interactions. The fiber-temperature and fiber-a_w interactions, influenced the degradation rate to a lesser degree due to the minimal effect fiber variability had on ascorbic acid stability. All temperature-a_w interactions were significantly different from one another except at 0.40 and 0.65 a_w at 37°C. This may be due to the lowered oxygen level dissolved in the moisture of the product at these higher temperatures.

The influence of the fiber-temperature interaction on the rate of ascorbic acid degradation was affected more

by temperature variability than by fiber variability (Appendix C; Figure 9). Likewise, the effect of the fiber-a_w interactions on ascorbic acid degradation was influenced more by the a_w than fiber variability (Appendix C; Figure 10, 11, 12). Although fiber had an effect on the degradation of the vitamin, as shown by the interactions of single variables, it was negligible when compared to effect of temperature and a_w.

CONCLUSION

Ascorbic acid stability was shown to fluctuate as a function of the induced variable conditions. Temperature and water activity variability, as affecting ascorbic acid stability has been well documented in previous studies (Lee and Labuza, 1975; Kirk et. al. (1977). This study has shown that water activity is not a perfect tool in analyzing ascorbic acid stability in a food product. Even at a given water activity, which is presently used as a measure of water availability and reactant mobility, differences in the half life values exist due to the presence of variable high molecular weight carbohydrates in the model systems. These differences in half life values would have been much more dramatic at a constant moisture content. Due to the significant differences between the fiber systems at a given water activity, one can justifiably state that water activity should not be the sole criteria upon which ascorbic acid stability is evaluated.

Utilizing various sources of fiber to increase the shelf-life of ascorbic acid might be justified at low a_w-low temperature storage conditions. However, this practice would be impractical at high a_w's stored at temperatures greater than 20°C. A more important aspect of this research might be the stabilization of water activity by fiber addition in order to increase moisture content and profit margin.

RESEARCH SUGGESTIONS

1) NMR studies could be utilized to determine whether the high water binding capacity fiber systems actually did reduce water mobility.
2) NMR could be used to adjust food systems to a given water mobility factor. The rate of ascorbic acid destruction should be measured as a function of water mobility within the various systems.
3) A correlation between water binding capacity (NMR), moisture content, and a_w, and their effect on the ascorbic acid degradation rate to provide a method with which to accurately predict ascorbic acid stability in a food product.
4) Use of highly unsaturated fat in a model system, with and without antioxidants, to demonstrate the possible effects of lipid oxidation on ascorbic acid stability and the relative effectiveness of various antioxidants.

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APPENDIX A

The Analysis of Variance was arrived by the following method. All half life values are derived from Table 4.

Sources of Variation: A_x = fiber

B_x = temperature

C_x = water activity

x indicates individual condition

Fiber Group Half Life Totals and Half Life Means (days)

	Totals	Mean (r=9)	pH
Food System #1 (A_1)	349	39	5.14
Food System #2 (A_2)	384	43	4.87
Food System #3 (A_3)	326	36	5.10
Food System #4 (A_4)	425	47	3.75
Food System #5 (A_5)	387	43	4.31
Food System #6 (A_6)	377	41	4.90

Temperature Group Half Life Totals and Half Life Means

	Totals	Mean (r=18)
Temperature B_1 (20°C)	1314	73
Temperature B_2 (30°C)	654	36
Temperature B_3 (37°C)	280	14

Water Activity Group Half Life Totals and Half Life Means

	Totals	Means (r=18)
Water Activity C_1 (.10 a_w)	1165	65
Water Activity C_2 (.40 a_w)	605	34
Water Activity C_3 (.65 a_w)	478	27

Fiber-Temperature Interaction Half Life

Totals and Half Life Means (days)

Fiber-Temperature	Totals	Means (r=3)
$(AB)_{1,1}$	205	68
$(AB)_{1,2}$	102	34
$(AB)_{1,3}$	42	14
$(AB)_{2,1}$	224	75
$(AB)_{2,2}$	112	37
$(AB)_{2,3}$	48	16
$(AB)_{3,1}$	186	62
$(AB)_{3,2}$	99	33
$(AB)_{3,3}$	41	14
$(AB)_{4,1}$	249	83
$(AB)_{4,2}$	121	40
$(AB)_{4,3}$	55	18
$(AB)_{5,1}$	229	76
$(AB)_{5,2}$	109	36
$(AB)_{5,3}$	49	16
$(AB)_{6,1}$	221	74
$(AB)_{6,2}$	111	37
$(AB)_{6,3}$	45	15

Fiber-Water Activity Interaction Half Life

Totals and Half Life Means (days)

Fiber-Water Activity	Totals	Mean (r=3)
$(AC)_{1,1}$	174	58
$(AC)_{1,2}$	94	31
$(AC)_{1,3}$	81	27
$(AC)_{2,1}$	205	68
$(AC)_{2,2}$	103	34
$(AC)_{2,3}$	76	25
$(AC)_{3,1}$	173	58
$(AC)_{3,2}$	80	27
$(AC)_{3,3}$	73	24
$(AC)_{4,1}$	219	73
$(AC)_{4,2}$	122	41
$(AC)_{4,3}$	84	28
$(AC)_{5,1}$	198	66
$(AC)_{5,2}$	107	36
$(AC)_{5,3}$	82	27
$(AC)_{6,1}$	196	65
$(AC)_{6,2}$	99	33
$(AC)_{6,3}$	82	27

Temperature-Water Activity Interaction Group

Totals and Means (days)

Temperature-Water Activity	Totals	Mean (r=3)
$(BC)_{1,1}$	670	112
$(BC)_{1,2}$	365	61
$(BC)_{1,3}$	279	47
$(BC)_{2,1}$	338	56
$(BC)_{2,2}$	173	29
$(BC)_{2,3}$	143	24
$(BC)_{3,1}$	159	27
$(BC)_{3,2}$	67	11
$(BC)_{3,3}$	56	9

Appendix B

ANALYSIS OF VARIANCE

$$SS_y = \sum_{i=1}^{54} y_i^2 - \left[(\sum^{54} y_1)^2/54 \right]$$

$$SS_y = 144,126 - 93,583 = 50,543$$

$$\text{C. F. (correction factor)} = (\sum^{54} y_1)^2/54 = 93,583$$

$$SS_A = (\sum_{i=1}^{6} A_i^2)\ /9 - \text{C.F.}$$

$$= \frac{848,056}{9} - 93,583 = 645$$

$$SS_B = \left(\sum_{i=1}^{3} B_i^2\right) /18 - C.F.$$

$$= \frac{(1314)^2 + (654)^2 + (280)^2}{18} - 93,583$$

$$= 30,457$$

$$SS_C = \left(\sum_{i=1}^{3} C_i^2\right) /18 - C.F.$$

$$= \frac{(1165)^2 + (605)^2 + (478)^2}{18} - 93,583$$

$$= 14,847$$

$$SS_{AB} = \left(\sum_{i=1}^{18} (AB)_i^2 /3\right) - C.F. - SS_A - SS_B$$

$$= \frac{374,872}{3} - 93,583 - 645 - 30,456$$

$$= 273$$

$$SS_{AC} = \left(\sum_{i=1}^{18} (AC)_i^2 /3\right) - C.F. - SS_A - SS_C$$

$$= \frac{327,960}{3} - 93,583 - 645 - 14,847$$

$$= 245$$

$$SS_{BC} = \left(\sum_{i=1}^{9} (BC)_i^2 /6\right) - C.F. - SS_B - SS_C$$

$$= \frac{857,494}{6} - 93,583 - 30,456 - 14,847$$

$$= 4,029$$

$$SS_E = SS_Y - (SS_A + SS_B + SS_C + SS_{AB} + SS_{AC} + SS_{BC})$$

$$= 50,543 - (645 + 30,456 + 14,847 + 273 + 245 + 4029)$$

$$= 48 = \text{Error SS}$$

The sum of squares was used in the Analysis of Variance. The half life means were used to draw conclusions using graphs and different statistical tests (Scheffe and Bonferroni t).

Energy of Activation as Calculated by Arrhenius Equation

Arrhenius Equation

$$\ln k = \ln A - \frac{E_a}{RT}$$

a_w	Fiber 1	Fiber 2	Fiber 3	Fiber 4	Fiber 5	Fiber 6
0.10	18.36	15.31	14.13	14.36	19.31	21.52
0.40	17.60	17.29	15.84	17.12	19.13	17.61
0.65	15.97	16.19	15.25	15.82	18.49	17.39

E_a values in K cals/mole.

Appendix C

Fiber-Temperature Interaction

Scheffe Test

Constant Temperature 20°C min. Sign. Diff.

$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{2,1}}) = 68.3 - 74.7 = -6.4$ 7.09

$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{3,1}}) = 68.3 - 62.0 = 6.3$ 7.09

$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{4,1}}) = 68.3 - 83.0 = -14.7$ 7.09

Continued

			min. sign. diff.
$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{5,1}})$	= 68.3 - 76.3	= -8.0	7.09
$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{6,1}})$	= 68.3 - 73.7	= -5.4	7.09
$(\bar{y}_{AB_{2,1}} - \bar{y}_{AB_{3,1}})$	= 74.7 - 62.0	= 14.7	7.09
$(\bar{y}_{AB_{2,1}} - \bar{y}_{AB_{4,1}})$	= 74.7 - 83.0	= -8.3	7.09
$(\bar{y}_{AB_{2,1}} - \bar{y}_{AB_{5,1}})$	= 74.7 - 76.3	= -1.6	7.09
$(\bar{y}_{AB_{2,1}} - \bar{y}_{AB_{6,1}})$	= 74.7 - 73.7	= 1.0	7.09
$(\bar{y}_{AB_{3,1}} - \bar{y}_{AB_{4,1}})$	= 62.0 - 83.0	= -21.0	7.09
$(\bar{y}_{AB_{3,1}} - \bar{y}_{AB_{5,1}})$	= 62.0 - 76.3	= -14.3	7.09
$(\bar{y}_{AB_{3,1}} - \bar{y}_{AB_{6,1}})$	= 62.0 - 73.7	= -11.7	7.09
$(\bar{y}_{AB_{4,1}} - \bar{y}_{AB_{5,1}})$	= 83.0 - 76.3	= 6.7	7.09
$(\bar{y}_{AB_{4,1}} - \bar{y}_{AB_{6,1}})$	= 83.0 - 73.7	= 9.3	7.09
$(\bar{y}_{AB_{5,1}} - \bar{y}_{AB_{6,1}})$	= 76.3 - 73.7	= 3.6	7.09

Fiber-Temperature Interaction

Scheffe Test

Constant Temperature $30^{o}C$ min. sign. diff.

$(\bar{y}_{AB_{1,2}} - \bar{y}_{AB_{2,2}})$	=	34.0 - 37.3	=	-3.3	7.09
$(\bar{y}_{AB_{1,2}} - \bar{y}_{AB_{3,2}})$	=	34.0 - 33.0	=	1.0	7.09
$(\bar{y}_{AB_{1,2}} - \bar{y}_{AB_{4,2}})$	=	34.0 - 40.3	=	-6.3	7.09
$(\bar{y}_{AB_{1,2}} - \bar{y}_{AB_{5,2}})$	=	34.0 - 36.3	=	-2.3	7.09
$(\bar{y}_{AB_{1,2}} - \bar{y}_{AB_{6,2}})$	=	34.0 - 37.0	=	-3.0	7.09
$(\bar{y}_{AB_{2,2}} - \bar{y}_{AB_{3,2}})$	=	37.3 - 33.0	=	4.3	7.09
$(\bar{y}_{AB_{2,2}} - \bar{y}_{AB_{4,2}})$	=	37.3 - 40.3	=	-3.0	7.09
$(\bar{y}_{AB_{2,2}} - \bar{y}_{AB_{5,2}})$	=	37.3 - 36.3	=	1.0	7.09
$(\bar{y}_{AB_{2,2}} - \bar{y}_{AB_{6,2}})$	=	37.3 - 37.0	=	0.3	7.09
$(\bar{y}_{AB_{3,2}} - \bar{y}_{AB_{4,2}})$	=	34.0 - 40.3	=	-6.3	7.09
$(\bar{y}_{AB_{3,2}} - \bar{y}_{AB_{5,2}})$	=	34.0 - 36.3	=	-2.3	7.09
$(\bar{y}_{AB_{3,2}} - \bar{y}_{AB_{6,2}})$	=	34.0 - 37.0	=	-3.0	7.09
$(\bar{y}_{AB_{4,2}} - \bar{y}_{AB_{5,2}})$	=	40.3 - 36.3	=	4.0	7.09
$(\bar{y}_{AB_{4,2}} - \bar{y}_{AB_{6,2}})$	=	40.3 - 37.0	=	3.3	7.09
$(\bar{y}_{AB_{5,2}} - \bar{y}_{AB_{6,2}})$	=	36.3 - 37.0	=	-0.7	7.09

Fiber-Temperature Interaction

Scheffe Test

Constant Fiber

				min. sign. diff.
Food System #1				
$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{1,2}})$	=	68.3 - 34.0	= 34.3	3.34
$(\bar{y}_{AB_{1,1}} - \bar{y}_{AB_{1,3}})$	=	68.3 - 14.0	= 54.3	3.34
$(\bar{y}_{AB_{1,2}} - \bar{y}_{AB_{1,3}})$	=	34.0 - 14.0	= 20.0	3.34
Food System #2				
$(\bar{y}_{AB_{2,1}} - \bar{y}_{AB_{2,2}})$	=	74.7 - 37.3	= 37.4	3.34
$(\bar{y}_{AB_{2,1}} - \bar{y}_{AB_{2,3}})$	=	74.7 - 16.0	= 58.7	3.34
$(\bar{y}_{AB_{2,2}} - \bar{y}_{AB_{2,3}})$	=	37.3 - 16.0	= 21.3	3.34
Food System #3				
$(\bar{y}_{AB_{3,1}} - \bar{y}_{AB_{3,2}})$	=	62.0 - 33.0	= 29.0	3.34
$(\bar{y}_{AB_{3,1}} - \bar{y}_{AB_{3,3}})$	=	62.0 - 13.7	= 48.3	3.34
$(\bar{y}_{AB_{3,2}} - \bar{y}_{AB_{3,3}})$	=	33.0 - 13.7	= 19.3	3.34
Food System #4				
$(\bar{y}_{AB_{4,1}} - \bar{y}_{AB_{4,2}})$	=	83.0 - 40.3	= 42.7	3.34
$(\bar{y}_{AB_{4,1}} - \bar{y}_{AB_{4,3}})$	=	83.0 - 18.3	= 64.7	3.34
$(\bar{y}_{AB_{4,2}} - \bar{y}_{AB_{4,3}})$	=	40.3 - 18.3	= 22.0	3.34

Food System #5

$(\bar{y}_{AB_{5,1}} - \bar{y}_{AB_{5,2}}) = 76.3 - 36.3 = 40.0 \qquad 3.34$

$(\bar{y}_{AB_{5,1}} - \bar{y}_{AB_{5,3}}) = 76.3 - 16.3 = 60.0 \qquad 3.34$

$(\bar{y}_{AB_{5,2}} - \bar{y}_{AB_{5,3}}) = 36.3 - 16.3 = 20.0 \qquad 3.34$

Food System #6

$(\bar{y}_{AB_{6,1}} - \bar{y}_{AB_{6,2}}) = 73.7 - 37.0 = 36.7 \qquad 3.34$

$(\bar{y}_{AB_{6,1}} - \bar{y}_{AB_{6,3}}) = 73.7 - 15.0 = 58.7 \qquad 3.34$

$(\bar{y}_{AB_{6,2}} - \bar{y}_{AB_{6,3}}) = 37.0 - 15.0 = 22.0 \qquad 3.34$

Fiber-Water Activity Interaction

Scheffe Test

Constant Water Activity 0.10

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{2,1}}) = 58.0 - 68.3 = -10.3 \qquad 7.09$

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{3,1}}) = 58.0 - 57.7 = 0.3 \qquad 7.09$

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{4,1}}) = 58.0 - 73.0 = -15.0 \qquad 7.09$

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{5,1}}) = 58.0 - 66.0 = -8.0 \qquad 7.09$

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{6,1}}) = 58.0 - 65.3 = -7.3 \qquad 7.09$

$(\bar{y}_{AC_{2,1}} - \bar{y}_{AC_{3,1}}) = 68.3 - 57.7 = 10.6 \qquad 7.09$

$(\bar{y}_{AC_{2,1}} - \bar{y}_{AC_{4,1}}) = 68.3 - 73.0 = -4.7 \qquad 7.09$

Continued

$(\bar{y}_{AC_{2,1}} - \bar{y}_{AC_{5,1}}) = 68.3 - 73.0 = -4.7 \qquad 7.09$

$(\bar{y}_{AC_{2,1}} - \bar{y}_{AC_{6,1}}) = 68.3 - 65.3 = 3.0 \qquad 7.09$

$(\bar{y}_{AC_{3,1}} - \bar{y}_{AC_{4,1}}) = 57.7 - 73.0 = -15.3 \qquad 7.09$

$(\bar{y}_{AC_{3,1}} - \bar{y}_{AC_{5,1}}) = 57.7 - 66.0 = -8.3 \qquad 7.09$

$(\bar{y}_{AC_{3,1}} - \bar{y}_{AC_{6,1}}) = 57.7 - 65.3 = -7.6 \qquad 7.09$

$(\bar{y}_{AC_{4,1}} - \bar{y}_{AC_{5,1}}) = 73.0 - 66.0 = 7.0 \qquad 7.09$

$(\bar{y}_{AC_{4,1}} - \bar{y}_{AC_{6,1}}) = 73.0 - 65.3 = 8.3 \qquad 7.09$

$(\bar{y}_{AC_{5,1}} - \bar{y}_{AC_{6,1}}) = 66.0 - 65.3 = 0.7 \qquad 7.09$

Fiber-Water Activity Interaction

Scheffe Test

Constant Water Activity 0.40 min. sign. diff.

$(\bar{y}_{AC_{1,2}} - \bar{y}_{AC_{2,2}}) = 31.3 - 34.3 = -3.0$ 7.09

$(\bar{y}_{AC_{1,2}} - \bar{y}_{AC_{3,2}}) = 31.3 - 26.7 = 4.6$ 7.09

$(\bar{y}_{AC_{1,2}} - \bar{y}_{AC_{4,2}}) = 31.3 - 40.7 = -9.4$ 7.09

$(\bar{y}_{AC_{1,2}} - \bar{y}_{AC_{5,2}}) = 31.3 - 35.7 = -4.3$ 7.09

$(\bar{y}_{AC_{1,2}} - \bar{y}_{AC_{6,2}}) = 31.3 - 33.0 = -1.7$ 7.09

$(\bar{y}_{AC_{2,2}} - \bar{y}_{AC_{3,2}}) = 34.3 - 26.7 = 7.6$ 7.09

$(\bar{y}_{AC_{2,2}} - \bar{y}_{AC_{4,2}}) = 34.3 - 40.7 = -6.4$ 7.09

$(\bar{y}_{AC_{2,2}} - \bar{y}_{AC_{5,2}}) = 34.3 - 35.7 = -1.4$ 7.09

$(\bar{y}_{AC_{2,2}} - \bar{y}_{AC_{6,2}}) = 34.3 - 33.0 = 1.3$ 7.09

$(\bar{y}_{AC_{3,2}} - \bar{y}_{AC_{4,2}}) = 26.7 - 40.7 = -14.0$ 7.09

$(\bar{y}_{AC_{3,2}} - \bar{y}_{AC_{5,2}}) = 26.7 - 35.7 = -9.0$ 7.09

$(\bar{y}_{AC_{3,2}} - \bar{y}_{AC_{6,2}}) = 26.7 - 33.0 = -6.3$ 7.09

$(\bar{y}_{AC_{4,2}} - \bar{y}_{AC_{5,2}}) = 40.7 - 35.7 = 5.0$ 7.09

$(\bar{y}_{AC_{4,2}} - \bar{y}_{AC_{6,2}}) = 40.7 - 33.0 = 7.7$ 7.09

$(\bar{y}_{AC_{5,2}} - \bar{y}_{AC_{6,2}}) = 35.7 - 33.0 = 2.7$ 7.09

Fiber-Water Activity Interactions

Scheffe Test

Constant Fiber

Food System #1 min. sign. diff.

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{1,2}}) = 58.0 - 31.3 = 26.7$ 3.34

$(\bar{y}_{AC_{1,1}} - \bar{y}_{AC_{1,3}}) = 58.0 - 27.0 = 31.0$ 3.34

$(\bar{y}_{AC_{1,2}} - \bar{y}_{AC_{1,3}}) = 31.3 - 27.0 = 4.3$ 3.34

Food System #2

$(\bar{y}_{AC_{2,1}} - \bar{y}_{AC_{2,2}}) = 68.3 - 34.3 = 34.0$ 3.34

$(\bar{y}_{AC_{2,1}} - \bar{y}_{AC_{2,3}}) = 68.3 - 25.3 = 43.0$ 3.34

$(\bar{y}_{AC_{2,2}} - \bar{y}_{AC_{2,3}}) = 34.3 - 25.3 = 9.0$ 3.34

Food System #3

$(\bar{y}_{AC_{3,1}} - \bar{y}_{AC_{3,2}}) = 57.7 - 27.7 = 30.0$ 3.34

$(\bar{y}_{AC_{3,1}} - \bar{y}_{AC_{3,3}}) = 57.7 - 24.3 = 33.4$ 3.34

$(\bar{y}_{AC_{3,2}} - \bar{y}_{AC_{3,3}}) = 27.7 - 24.3 = 3.4$ 3.34

Food System #4

$(\bar{y}_{AC_{4,1}} - \bar{y}_{AC_{4,2}}) = 73.0 - 40.7 = 32.3$ 3.34

$(\bar{y}_{AC_{4,1}} - \bar{y}_{AC_{4,3}}) = 73.0 - 28.0 = 45.0$ 3.34

$(\bar{y}_{AC_{4,2}} - \bar{y}_{AC_{4,3}}) = 40.7 - 28.0 = 12.7$ 3.34

Food System #5

$(\bar{y}_{AC_{5,1}} - \bar{y}_{AC_{5,2}}) = 66.0 - 35.7 = 30.3 \qquad 3.34$

$(\bar{y}_{AC_{5,1}} - \bar{y}_{AC_{5,3}}) = 66.0 - 27.3 = 38.7 \qquad 3.34$

$(\bar{y}_{AC_{5,2}} - \bar{y}_{AC_{5,3}}) = 35.7 - 27.3 = 8.4 \qquad 3.34$

Food System #6

$(\bar{y}_{AC_{6,1}} - \bar{y}_{AC_{6,2}}) = 65.3 - 33.0 = 32.3 \qquad 3.34$

$(\bar{y}_{AC_{6,1}} - \bar{y}_{AC_{6,3}}) = 65.3 - 27.3 = 36.0 \qquad 3.34$

$(\bar{y}_{AC_{6,2}} - \bar{y}_{AC_{6,3}}) = 33.0 - 27.3 = 5.7 \qquad 3.34$

Temperature-Water Activity Interaction

Scheffe Test

Constant Temperature 20°C min. sign. diff.

$(\bar{y}_{BC_{1,1}} - \bar{y}_{BC_{1,2}}) = 111.7 - 60.8 = 46.0 \qquad 2.363$

$(\bar{y}_{BC_{1,1}} - \bar{y}_{BC_{1,3}}) = 111.7 - 46.5 = 60.3 \qquad 2.363$

$(\bar{y}_{BC_{1,2}} - \bar{y}_{BC_{1,3}}) = 60.8 - 46.5 = 14.3 \qquad 2.363$

Constant Temperature 30°C

$(\bar{y}_{BC_{2,1}} - \bar{y}_{BC_{2,2}}) = 56.3 - 28.8 = 27.5 \qquad 2.363$

$(\bar{y}_{BC_{2,1}} - \bar{y}_{BC_{2,3}}) = 56.3 - 23.8 = 32.5 \qquad 2.363$

$(\bar{y}_{BC_{2,2}} - \bar{y}_{BC_{2,3}}) = 28.8 - 23.8 = 5.0 \qquad 2.363$

Constant Temperature 37°C — min. sign. diff.

$(\bar{y}_{BC_{3,1}} - \bar{y}_{BC_{3,2}}) = 26.5 - 11.2 = 15.3$ — 2.363

$(\bar{y}_{BC_{3,1}} - \bar{y}_{BC_{3,3}}) = 26.5 - 9.3 = 17.2$ — 2.363

$(\bar{y}_{BC_{3,2}} - \bar{y}_{BC_{3,3}}) = 11.2 - 9.3 = 1.9$ — 2.363

Temperature-Water Activity Interaction

Scheffe Test

Constant Water Activity 0.10

$(\bar{y}_{BC_{1,1}} - \bar{y}_{BC_{2,1}}) = 111.7 - 56.3 = 55.4$ — 2.363

$(\bar{y}_{BC_{1,1}} - \bar{y}_{BC_{3,1}}) = 111.7 - 26.5 = 85.2$ — 2.363

$(\bar{y}_{BC_{2,1}} - \bar{y}_{BC_{3,1}}) = 56.3 - 26.5 = 30.3$ — 2.363

Constant Water Activity 0.40

$(\bar{y}_{BC_{1,2}} - \bar{y}_{BC_{2,2}}) = 60.8 - 28.8 = 32.0$ — 2.363

$(\bar{y}_{BC_{1,2}} - \bar{y}_{BC_{3,2}}) = 60.8 - 11.2 = 49.6$ — 2.363

$(\bar{y}_{BC_{2,2}} - \bar{y}_{BC_{3,2}}) = 28.8 - 11.2 = 17.6$ — 2.363

Constant Water Activity 0.65

$(\bar{y}_{BC_{1,3}} - \bar{y}_{BC_{2,3}}) = 46.5 - 23.8 = 22.7$ — 2.363

$(\bar{y}_{BC_{1,3}} - \bar{y}_{BC_{3,3}}) = 46.5 - 9.3 = 35.2$ — 2.363

$(\bar{y}_{BC_{2,3}} - \bar{y}_{BC_{3,3}}) = 23.8 - 9.3 = 14.5$ — 2.363