EXTRACTS OF GARDEN VEGETABLES AS SOURCES OF NUTRITION FOR VARIOUS MICROORGANISMS

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CHAPTER I

INTRODUCTION

Since the beginning of time, both man and lower animals have relied upon plant life for their subsistence. Certain plants have been domesticated, and today an important part of man's diet is composed of these. Such plants are commonly called garden vegetables and contain many of the vital nutrients which are necessary for the survival of the human race. Some studies have been made on many of these and some of their constituents have been revealed. Some of these vegetables are presently used in bacteriological culture media; however, they are not used alone, but in addition to other nutritional materials. This study was undertaken in order to determine whether the extracts of common garden vegetables could be incorporated into simple, economical culture media which might be used for the growth and cultivation of at least some of the more commonly used microorganisms.

This investigation included the collecting, extracting, and testing of the vegetable extracts for microorganism nutritive value, in vitro, of twenty-five common garden vegetables and five mixtures containing various vegetable

extracts. For testing purposes thirty species of microorganisms were used. In this group of test organisms there were ten species of gram-positive and ten species of gram-negative bacteria, two species of molds or higher fungi, three species of yeasts, and five species of actinomycetes. The extracts of these common vegetables were incorporated into simple culture media. A culture of each species of organisms was transplanted thereon, incubated, the amount of growth determined, and the results recorded.

Review of Literature

One of the most significant factors influencing bacterial growth is the presence of amino acids. Steinman and Eagle (1948) produced a medium upon which the nonpathogenic Reiter strain of treponeme could be cultivated. This medium consisted of arginine, acetic acid, a sulfahydryl-containing compound, crystallized serum albumin, and Brewer's thioglycolate medium. The essential component in the latter was an enzymatic digest of casein.[1] Later Steinman, Eagle, and Oyana (1952) devised a medium which did not require the enzymatic casein hydrolysate. This medium consisted of amino acids, vitamins, purines, pyrimidines, carbohydrates,

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[1] H. Eagle and H. G. Steinman, "The Nutritional Requirements of Treponemata," Journal of Bacteriology, LVI (1948), 163-176.

inorganic ions, and crystallized albumin.[2] Drew and Mueller (1951) investigated the amino acid requirements of Cornybacterium diphtheriae, Toronto strain of Park-Williams number eight. They showed that excellent growth and toxin production could be obtained on a medium composed of eight amino acids, ammonium sulfate, trace metals, inorganic salts, four accessory growth factors and maltose.[3] Whiteside-Carlson and Rosano (1951) determined the amino acid, vitamin, purine, and pyrimidine, and mineral requirements of Leuconostoc dextranicum, strain "elai" for growth in sucrose, glucose, and fructose media. Essential amino acids were glutamic acid, valine, threonine, tryptophane, and histidine.[4] Pratt (1952) described a medium containing glucose, Tween 80 (polyoxyethelene derivative of sorbitan monooleate), and acid hydrolyzed casein which produced rapid growth of Mycobacterium phlei. In the absence of Tween 80 the organism grew very slowly.[5] Hassinen and others (1951) made a study

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[2]H. G. Steinman, H. Eagle, and V. I. Oyana, "The Nutritional Requirements of Treponemata," Journal of Bacteriology, LXIV (1952), 265-269.

[3]Ruth M. Drew and J. Howard Mueller, "A Chemically Defined Medium Suitable for the Production of Higher Titer Diphtherial Toxin," Journal of Bacteriology, LXII (1951), 549-559.

[4]Virginia Whiteside-Carlson and Carmen L. Rosano, "The Nutritional Requirements of Leuconostoc Dextranicum for Growth and Dextran Synthesis," Journal of Bacteriology, LXII (1951), 583-589.

[5]Darrell Pratt, "Nutrition of Mycobacterium Phlei," Journal of Bacteriology, LXIV (1952), 659-665.

of the mineral nutritional requirements of four strains of Lactobacillus bifidus isolated from the stools of breast-fed infants. They found that the strains would grow on a simplified medium comprised of lactose, buffers, minerals, cysteine, ammonium salts, and the vitamins, biotin and calcium pantothenate.[6]

Katznelson and Lochhead (1952) described the growth factor requirements of halophilic bacteria. The growth factor requirements of organisms ranging in optimal sodium chloride requirements from four per cent to twenty-five per cent, including nonchromogenic forms, were studied. A species of Vibrio (No. 756), Micrococcus species (No. 732), and Sarcina species (No. $63N_2$) represented the colorless organisms, and Pseudomonas salinaria, Pseudomonas cutirubra, and Sarcina littoralis represented the colored forms. All were tested in a basal medium containing vitamin-free casamino acids, salts, citrate, and succinate. The Vibrio species grew well in the basal medium but was stimulated by ribonucleic acids as its pyrimidines, uracil and cytosine. The Micrococcus species required thiamin for growth and was stimulated by purines and pyrimidines. Sarcina species required thiamin, pantothenic acid, nicotinic acid, and biotin for growth and was stimulated by folic and p-aminobenzoic acids, uracil, choline, and pyridoxal. Growth

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[6]J. D. Hassinen and others, "The Minimal Nutritional Requirements of Lactobacillus Bifidus," Journal of Bacteriology, LXII (1951), 771-777.

factors did not stimulate the colored halophiles but yeast extract improved growth of the organisms.[7] Clark (1949) showed that the nitrogen requirements of Clostridium thermosaccharolyticum, when grown in a synthetic basal medium, could be supplied by ammonium sulfate, methionine, cystine, and a combination of valine, leucine, and isoleucine.[8] Gerhardt, Tucker, and Wilson (1950) revealed that l--or--dl-glutamic acid, l--or--dl-asparagine, and l-histidine supported good growth of Brucella abortus, strain 19, when supplied singly as the nitrogen source in a chemically defined medium.[9] Pomper (1952) disclosed that mutant number (62-28) of Saccharomyces cerevisiae required pantothenic acid, p-aminobenzoic acid, methionine, adenine, and histidine at pH 6.8. When the pH is lowered from 6.8 to 5.5 the adenine and histidine are not needed. Methionine, adenine, histidine, and pantothenic acid were replaced by increased concentrations of p-aminobenzoic acid.

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[7]H. Katznelson and A. G. Lochhead, "Growth Factor Requirements of Halophilic Bacteria," Journal of Bacteriology, LXIV (1952), 97-103.

[8]F. M. Clark, "Some Nitrogen Requirements of Clostridium Thermosaccharolyticum," Journal of Bacteriology, LVII (1949), 465-471.

[9]Phillip Gerhardt, L. A. Tucker, and J. B. Wilson, "The Nutrition of Brucellae: Utilization of Single Amino Acids for Growth," Journal of Bacteriology, LIX (1950), 777-782.

P-aminobenzoic acid was able to be replaced by tyrosine and certain other para substituted acids.[10]

A second significant growth factor necessary for microorganisms is the presence of various metallic ions. MacLeod (1951) demonstrated that Streptococcus fecalis, strain R, required both Mg^{++} and Mn^{++} for growth. Other related ions tested were unable to replace Mg^{++} and Mn^{++} in the nutrition of the organism.[11] Shanker and Bard (1952) described a metal-deficient complex medium which was employed to determine the major metallic ion growth requirements of Clostridium perfringens. For optimum growth, Ca^{++}, Mg^{++}, Fe^{++}, Na^{++}, and K^{+} are required but not Zn^{++}, Mn^{++}, Co^{++}, and Cu^{++}. None of the latter metallic ions can replace those required for growth.[12] Charney, Fisher, and Hagerty (1951) showed that manganese stimulates sporulation of several species of the genus Bacillus.[13]

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[10]Seymour Pomper," A pH-Sensitive Multiple Mutant of Saccharomyces Cerevisiae," Journal of Bacteriology, LXIV (1952), 353-361.

[11]Robert A. MacLeod, "Further Mineral Requirements of Streptococcus Fecalis," Journal of Bacteriology, LXII (1951), 337-345.

[12]K. Shanker and R. C. Bard, "The Effect of Metallic Ions on the Growth and Morphology of Clostridium Perfringens," Journal of Bacteriology, LXIII (1952), 279-289.

[13]Jesse Charney, W. P. Fisher, and C. P. Hagerty, "Manganese As An Essential Element for Sporulation in the Genus Bacillus," Journal of Bacteriology, LXII (1951), 145-148.

Some investigators have exhibited the need for various organic acids in the metabolism of many microorganisms. Johnson and Cohn (1952) showed that when malic and fumaric acids were added to a basal medium for Escherichia coli at levels up to five hundred micromoles per ten cubic centimeters of media, growth was markedly stimulated.[14] Poe and Charkey (1949) showed the effect of boric acid on the growth of cultures of the genera Escherichia and Aerobacter.[15] Ackart and Murray (1951) modified the Dubos medium by adding succinic acid as a source of carbon for the growth of Mycobacterium avium.[16]

Still another group of food materials, principally organic compounds, have been found necessary, in minute amounts only, for growing microorganisms. These are referred to as growth accessory substances. Lochhead and Thexton (1952) reported that for a marked proportion of the indigenous bacteria of soil that depend upon essential growth factors supplied by soil extract, the growth-promoting

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[14]B. Connor Johnson and Eva M. Cohn, "Effect of Certain Acids of the Tricarboxylic Acid Cycle on the Growth of Escherichia Coli," Journal of Bacteriology, LXIII (1952), 735-742.

[15]Charles F. Poe and Lowell W. Charkey, "A Study of Boric Acid Media for the Separation of Escherichia and Aerobacter," Journal of Bacteriology, LVII (1949), 386-387.

[16]W. B. Ackart and T. J. Murray, "A Medium for the Rapid Cultivation of Mycobacterium Avium," Journal of Bacteriology, LXII (1951), 75-79.

effect of the latter could be replaced by vitamin B_{12}.[17] Starr (1949) determined the mineral nutritive requirements of six species of gram-positive phytopathogenic coryne-bacteria. All species required obligate additions to the ammonium chloride, glucose, and salts basal medium, as follows: C. fascians, thiamine, C. flaccumfaciens and C. poinsettiae, amino acids, biotin, pantothenate, and thiamine; C. insidiosum, C. michiganense, and C. sepedonicum, amino acids, biotin, nicotinic acid and thiamine.[18] Delwiche (1949) reported that the B vitamin requirements of twenty-five different strains of the genus Propionibacterium on synthetic media were relatively simple. The most demanding cultures required only pantothenic acid, biotin, and either thiamine or p-aminobenzoic acid.[19] Cleverdon, Pelczar, and Doetsch (1949) reported that twelve stenothermophiles grew well in serial transfer, at fifty-five degrees centigrade and sixty-five degrees centigrade, in a casein hydrolysate medium when supplied biotin, four one-hundredths milligram per milliliter, and niacin and thiamin each one milligram per milliliter.[20] Smith and Douglas (1950) found that the

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[17]A. G. Lochhead and R. H. Thexton, "Qualitative Studies of Soil Microorganisms," Journal of Bacteriology, LXIII (1952), 219-226.

[18]Mortimer P. Starr, "The Nutrition of Phytopathogenic Bacteria," Journal of Bacteriology, LVII (1949), 253-257.

[19]Eugene A. Delwiche, "Vitamin Requirements of the Genus Propionbacterium," Journal of Bacteriology, LVIII (1949), 395-398.

[20]Robert C. Cleverdon, Michael J. Pelczar, Jr., and Raymond N. Doetsch, "The Vitamin Requirements of Stenothermophilic Aerobic Sporogenous Bacilli," Journal of Bacteriology, LVIII (1949), 523-526.

maximum growth of Clostridium bifermentans in a medium composed of "vitamin-free" acid-hydrolyzed casein, glucose, and inorganic salts was dependent upon added biotin, nicotinic acid or amide, pantothenic acid, and pyridoxal or pyridoxamine. Biotin was not required by some strains and pyridoxamine was not required by others.[21] Gaines and Stahly (1943) used a medium in which all constituents except casein hydrolysate were chemically defined. They found that thiamin, calcium pantothenate, and nicotinic acid were essential for the growth of Leuconostoc mesenteroides 535 and that pyridoxin exerted a stimulatory effect. Biotin also stimulated growth of the organism.[22]

Carbohydrates are essential to organisms and can be utilized as the sole source of carbon. Liu (1952) showed that Pseudomonas aeruginosa has a definite biochemical pattern. This organism is able to utilize glucose, galactose, fructose, mannitol, trehalose, and glycerol, as the sole source of carbon for growth and to produce acid from these substances. The organism is unable to utilize arabinose, xylose, and mannose as the sole source of carbon for growth, although acid is produced rapidly from these.[23]

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[21]Louis DeSpain Smith and Howard C. Douglas, "Factors Necessary for Maximum Growth of Clostridium Bifermentans," Journal of Bacteriology, LX (1950), 9-15.

[22]Sidney Gaines and Grant L. Stahly, "The Growth Requirements of Leuconostoc Mesenteroides and Preliminary Studies on Its Use as an Assay Agent for Several Members of the Vitamin B Complex," Journal of Bacteriology, XXXXVI 441-449.

[23]Pinghm Liu, "Utilization of Carbohydrates by Pseudomonas Aeruginosa," Journal of Bacteriology, LXIV (1952), 773-781.

Kitay and Snell (1950) made a survey of additional nutritive requirements of twenty-eight cultures of lactic acid bacteria previously reported unable to grow in a medium of known composition. These included Lactobacillus delbrueckii, Lactobacillus acidophilus, Lactobacillus bulgaricus, Lactobacillus helveticus, Lactobacillus lactis, Lactobacillus leichmannii, and Leuconostoc citrovorum. All of these cultures except Lactobacillus lactis, and Leuconostoc citrovorum required oleic acid or other unsaturated fatty acids for growth. Eighteen of the twenty-eight cultures required thymidine, other desoxyribosides, or vitamin B_{12} for growth.[24]

The Problem

The purpose of this investigation is to determine the possible presence of nutrient constituents in the extracts of several common garden vegetables as sources of useful, satisfactory, and economical culture media which can be used in biological laboratories, both on the secondary and higher education levels. The problem has consisted of, first, the collection and preparation of a number of different kinds of garden vegetables; second, the procuring of the extracts from these; third, the preparation of culture media using the vegetable extracts as the sources of the nutritional requirements for the growth of twenty bacterial organisms, two

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[24]Estelle Kitay and Esmond E. Snell, "Some Additional Nutritional Requirements of Certain Lactic Acid Bacteria," Journal of Bacteriology, LX (1950), 49-56.

species of molds, three species of yeasts, and five strains of actinomycetes; and fourth, the evaluation of these extracts, in terms of growth supported, as possible nutrient materials suitable for cultivation of the test microorganisms and possibly for others, similar to these.

CHAPTER II

GARDEN VEGETABLES USED, TEST MICROORGANISMS, AND LABORATORY WORK

Garden Vegetables Used

The names of the garden vegetables used in this investigation are listed in Table 1. The scientific names are in alphabetical order and are followed by the common names. The physical state, fresh or frozen, of each vegetable obtained is also given in the same table. All of the vegetables are ones commonly consumed by people. Since the laboratory work of this investigation was done during the spring and summer months, fresh vegetables were easily obtained. Some frozen ones were used, however.

Test Microorganisms

The microorganisms used in this investigation are listed in Table 2. The ten gram-positive bacterial organisms are listed first and these are followed by the ten gram-negative ones. The two species of molds, three species of yeasts, and five strains of actinomycetes are listed in that order. The five strains of actinomycetes, designated as X, 2, 4, 5, and 44, were obtained from isolated cultures used in a previous investigation by another graduate student working in

the same laboratory. All of these organisms were cultivated on Tryptose agar slants and in Tryptose broth. The vegetable

TABLE 1

VEGETABLES FROM WHICH EXTRACTS WERE OBTAINED

Scientific Name*	Common Name	Physical State
Allium cepa	Onion	Fresh
Apium graveolens	Celery	Fresh
Asparagus officinalis	Asparagus	Frozen
Beta cicla	Swiss chard	Fresh
Beta crassa	Beet	Fresh
Brassica oleracea	Cabbage	Fresh
Brassica oleracea var. botrytis I	Broccoli (leaves)	Fresh
Brassica oleracea var. botrytis II	Cauliflower	Frozen
Brassica rapa	Turnip	Fresh
Capsicum annuum	Green pepper	Fresh
Cucumis sativus	Cucumber	Fresh
Cucurbita maxima	Squash (yellow)	Fresh
Daucus carota	Carrot	Fresh
Hibiscus esculentus	Okra	Fresh
Lactuca sativa	Lettuce	Fresh
Lycopersicum esculentum	Tomato	Fresh
Petroselinum hortense	Parsley	Fresh
Phaseolus lunatus	Lima bean	Frozen
Phaseolus vulgaris	Green bean	Frozen
Pisum sativum	Green pea	Frozen
Rheum rhaponticum	Rhubarb	Fresh
Solanum melongena	Egg plant	Fresh
Spinacia oleracea	Spinach	Frozen
Vigna sinensis	Blackeyed pea	Frozen
Zea saccharata	Corn (yellow)	Frozen

*L. H. Bailey, The Standard Cyclopedia of Horticulture, (3 vols.), 1935.

extract agar plates were inoculated from the Tryptose broth cultures, whereas the vegetable extract broth was inoculated from the Tryptose agar slants. Each organism was transferred

semi-monthly to new stock culture medium in order to assure consistent growth reactions on the various test extract media.

TABLE 2

TEST MICROORGANISMS

Organism	Morphology
Bacillus cereus Frankland*	Bacillus
Bacillus megatherium De Bary	Bacillus
Bacillus mycoides Flugge	Bacillus
Bacillus subtilis Cohn	Bacillus
Gaffkya tetragena (Gaffkya) Trevisan	Coccus
Micrococcus pyogenes var. albus Rosenbach	Coccus
Micrococcus pyogenes var. aureus Rosenbach	Coccus
Micrococcus roseus Flugge	Coccus
Micrococcus citreus Migula	Coccus
Mycobacterium smegmatis (Trevisan) Chester	Bacillus
Aerobacter aerogenes (Kruse) Beijerinck	Bacillus
Alkaligenes fecalis Castellani and Chalmers	Bacillus
Escherichia coli (Migula) Castellani	Bacillus
Escherichia communior (Topley) Yale	Bacillus
Klebsiella pneumoniae (Schroeter) Trevisan	Bacillus
Neisseria catarrhalis (Frosch) Holland	Coccus
Pseudomonas aeruginosa (Schroeter) Migula	Bacillus
Pseudomonas fluorescens Migula	Bacillus
Proteus vulgaris Hauser	Bacillus
Serratia marcescens Bizio	Bacillus
Aspergillus niger (Michx) Tiegh**	Mold
Penicillium notatum Link	Mold
Saccharomyces cerevisiae (Meyer) Hansen	Yeast
Saccharomyces ellipsoideus (Meyer) Hansen	Yeast
Torula histolytica Turpin	Yeast
Actinomycete X	Actinomycete
Actinomycete 2	Actinomycete
Actinomycete 4	Actinomycete
Actinomycete 5	Actinomycete
Actinomycete 44	Actinomycete

*D. Bergey, et al., Manual of Determinative Bacteriology, 6th edition, 1948.

**G. C. Ainsworth and G. R. Bisby, A Dictionary of the Fungi, 1945.

Laboratory Work

Preparation and use of the vegetable extract agar.--One pound of each vegetable was weighed, washed thoroughly if necessary, and ground into small particles in a food grinder. The ground portion, along with the juice produced in the grinding procedure, was then placed in a two liter flask. To this seven hundred cubic centimeters of distilled water was added, the mixture heated for thirty minutes, and allowed to cool. The extract was decanted from the solid vegetable residue and filtered free of all vegetable particles. The extract was then made up to the original volume of seven hundred cubic centimeters. Seven and one-half grams of Bacto-Agar was added to five hundred cubic centimeters of the extract in a clean flask. This was stoppered with a clean cotton stopper and sterilized in the autoclave for fifteen minutes at one hundred twenty degrees centigrade and fifteen pounds pressure per square inch. When sufficiently cooled, fifteen cubic centimeters of the vegetable extract agar was poured into each of thirty-one sterile petri dishes. The agar was allowed to cool, and the plates were labeled properly. Each plate was inoculated with one of the thirty test organisms listed in Table 2. One plate of medium was used as a control. The plates were incubated for forty-eight hours at thirty-seven degrees centigrade and observed for results. The pH of each vegetable extract was determined by the use of a pH

meter. No adjustments were made in the natural pH. These values are listed in Table 3.

Preparation and use of extract broth.--The remaining two hundred cubic centimeters of each vegetable extract was placed in thirty-one test tubes in five to seven cubic centimeter aliquots. The tubes were stoppered and autoclaved for

TABLE 3

pH OF VEGETABLE EXTRACTS

Vegetable	pH
Allium cepa	7.22
Apium graveolens	7.22
Asparagus officinalis	7.30
Beta cicla	6.00
Beta crassa	6.24
Brassica oleracea	6.20
Brassica oleracea var. botrytis I	5.00
Brassica oleracea var. botrytis II	5.87
Brassica rapa	7.08
Capsicum annuum	6.93
Cucumis sativus	4.02
Cucurbita maxima	5.70
Daucus carota	6.00
Hibiscus esculentus	8.07
Lactuca sativa	7.38
Lycopersicum esculentum	6.24
Petroselinum hortense	7.42
Phaseolus lunatus	7.58
Phaseolus vulgaris	5.30
Pisum sativum	5.68
Rheum rhaponticum	3.95
Solanum melongena	6.90
Spinacia oleracea	7.23
Vigna sinensis	5.28
Zea saccharata	7.10

fifteen minutes at one hundred twenty degrees centigrade and fifteen pounds pressure per square inch. Upon cooling, each

of the tubes was inoculated with one of the thirty test organisms. One tube was kept as a control. The broth cultures were incubated for forty-eight hours at thirty-seven degrees centigrade and observed for results.

Preparation and use of vegetable extract mixtures.--The vegetable extract mixtures were prepared by using aliquots of excess extracts not needed in the previous experiments. Ordinarily one pound of vegetable was use per seven hundred cubic centimeters of water. However, those used in mixtures

TABLE 4

COMPOSITION OF VEGETABLE EXTRACT MIXTURES

Mixture	Contents of Mixture	Cubic Centimeters Used
Number 1	Beta cicla	100
	Beta crassa	100
	Brassica oleracea var. botrytis I	100
	Cucumis sativus	100
	Distilled water	300
Number 2	Pisum sativum	200
	Rheum rhaponticum	100
	Distilled water	400
Number 3	Vigna sinensis	250
	Phaseolus vulgaris	250
	Distilled water	200
Number 4	Pisum sativum	50
	Vigna sinensis	50
	Rheum rhaponticum	50
	Mixture Number 2	50
	Mixture Number 3	50
	Distilled water	350
Number 5	Brassica oleracea var. botrytis II	150
	Zea saccharata	300
	Distilled water	250

were made up in larger quantities in order to produce the excess for mixtures and still retain the one pound of vegetables to seven hundred cubic centimeters of water ratio. The vegetable extracts and the amounts used in the various mixtures are listed in Table 4. The extract broth and extract agar mixtures were made up in the same manner as the individual vegetable extract media. In addition, the inoculation procedure, incubation, and observation of results were the same.

CHAPTER III

EXPERIMENTAL DATA AND DISCUSSION

Experimental Data

After an incubation period of forty-eight hours, the extract agar plates and broths were observed and the results recorded. The growth on the agar plates was evaluated in terms of the number and size of colonies. The criterion for evaluating plates with spreading growth was based upon the fraction of the plate covered by this growth. In evaluating the resulting growth in the extract broth cultures, each of the inoculated tubes was compared with the control tube. The degree of turbidity in each of the thirty inoculated tubes, when compared with the control, was used as the basis of evaluation.

The symbols used in recording the growth on the agar plates and in the broth cultures are shown in Table 5. The results were recorded in plus (+) signs which range from barely perceptible growth, one plus (+), to heavy growth, three plus (+++). The nutritional value of the vegetable extracts for the microorganisms used in this investigation are shown in Tables 6 through 35.

Table 6 shows the nutritive value of the two Allium cepa extra media. Bacillus mycoides, Bacillus subtilis, Aerobacter aerogenes, Klebsiella pneumoniae, Pseudomonas fluorescens, and Serratia marcescens produced excellent growth on both media. Bacillus megatherium, Micrococcus roseus, Alkaligenes fecalis, Escherichia coli, Escherichia communior, and Proteus vulgaris grew only slightly. All of

TABLE 5

MEANING OF SYMBOLS USED IN EVALUATION OF VEGETABLE NUTRIENTS FOR THIRTY TEST MICROORGANISMS

Symbol	Meaning
-	. . . . . No growth observed; indicates no nutritive value.
+	. . . . . Barely perceptible growth; less than ten small colonies or fewer larger ones.
++	. . . . . Good growth; ten to twenty-five small colonies or fewer larger ones.
+++	. . . . . Heavy growth; more than twenty-five small colonies or fewer larger ones.

the yeasts and molds grew in these media. Torula histolytica, however, grew only slightly in the extract broth. Actinomycete 44 grew the most luxuriantly of any of the members of this group. Several other bacterial species grew moderately both upon the vegetable extract agar medium and in the vegetable extract broth.

An observation of Table 7 reveals the nutritive value of the extract media of Apium graveolens. Gaffkya tetragena,

TABLE 6

NUTRITIVE VALUE OF ALLIUM CEPA
EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	+
Bacillus megatherium	+	+
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	++
Micrococcus pyogenes var. albus	++	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	+	+
Micrococcus citrous	-	+
Mycobacterium smegmatis	-	+++
Aerobacter aerogenes	+++	+++
Alkalegenes fecalis	+	+
Escherichia coli	+	+
Escherichia communior	+	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	+
Pseudomonas aeruginosa	++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	+	+
Serratia marcescens	+++	+++
Aspergillus niger	++	+++
Penicillium notatum	+	+
Saccharomyces cerevisiae	+++	+
Saccharomyces ellipsoideus	++	+
Torula histolytica	-	+
Actinomycete X	++	++
Actinomycete 2	+	+
Actinomycete 4	+	+
Actinomycete 5	+	++
Actinomycete 44	+++	+++

Micrococcus citreus, Neisseria catarrhalis, Micrococcus pyogenes var. albus, Micrococcus roseus, and Torula histolytica did not grow at all on either of these preparations. Aspergillus niger, Bacillus subtilis, and Aerobacter

aerogenes grew very well in both of these, whereas Actinomycete 5 and Actinomycete 44 grew only moderately.

TABLE 7

NUTRITIVE VALUE OF *APIUM GRAVEOLENS* EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	++
Bacillus megatherium	+	+++
Bacillus mycoides	++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	+	-
Micrococcus roseus	-	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	+	++
Escherichia coli	++	+
Escherichia communior	+	-
Klebsiella pneumoniae	++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	++	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	+	-
Serratia marcescens	+	+++
Aspergillus niger	+++	+++
Penicillium notatum	++	-
Saccharomyces cerevisiae	++	-
Saccharomyces ellipsoideus	++	-
Torula histolytica	-	-
Actinomycete X	+	+++
Actinomycete 2	+	+
Actinomycete 4	+	-
Actinomycete 5	++	++
Actinomycete 44	++	+++

Table 8 shows the nutritive value of *Asparagus officinalis* extract media. Eight species of bacterial organisms

TABLE 8

NUTRITIVE VALUE OF *ASPARAGUS OFFICINALIS* EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	+
Bacillus megatherium	++	+
Bacillus mycoides	-	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	+	+
Micrococcus roseus	-	+
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	+	-
Escherichia coli	++	+
Escherichia communior	++	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	++	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	+	-
Serratia marcescens	+	+
Aspergillus niger	+	++
Penicillium notatum	+	+
Saccharomyces cerevisiae	++	+
Saccharomyces ellipsoideus	++	+
Torula histolytica	+	-
Actinomycete X	+	-
Actinomycete 2	-	-
Actinomycete 4	-	-
Actinomycete 5	+	-
Actinomycete 44	+	+

showed medium to heavy growth while *Micrococcus pyogenes* var. *albus*, *Micrococcus pyogenes* var. *aureus*, and *Serratia marcescens* showed very scant growth. Two species of yeasts, *Saccharomyces cerevisiae* and *Saccharomyces ellipsoideus*,

showed some growth, while the molds, Aspergillus niger and Penicillium notatum, grew fair to moderately.

The nutritive value of the extract media of Beta cicla is shown in Table 9. Five of the test organisms failed to

TABLE 9

NUTRITIVE VALUE OF BETA CICLA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	++
Bacillus megatherium	+	++
Bacillus mycoides	+	++
Bacillus subtilis	+++	++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	++	+
Micrococcus pyogenes var. aureus	++	++
Micrococcus roseus	++	+
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	-
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	++	+++
Escherichia communior	-	-
Klebsiella pneumoniae	-	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	++	++
Proteus vulgaris	+++	-
Serratia marcescens	+++	++
Aspergillus niger	+++	+++
Penicillium notatum	++	++
Saccharomyces cerevisiae	-	+++
Saccharomyces ellipsoideus	-	++
Torula histolytica	+	++
Actinomycete X	-	+++
Actinomycete 2	+	+++
Actinomycete 4	+	+++
Actinomycete 5	-	+++
Actinomycete 44	+	+++

grow in either of the two extract media. These were Gaffkya tetragena, Micrococcus citreus, Mycobacterium smegmatis, Alkaligenes fecalis, and Neisseria catarrhalis. Relatively consistent good growth of organisms, except the ones mentioned previously, was exhibited in the extract broth medium. Bacillus subtilis, Aerobacter aerogenes, Pseudomonas aeruginosa, Proteus vulgaris, Serratia marcescens, and Aspergillus niger grew luxuriantly on the beet extract agar.

The nutritive value of Beta crassa extract media is shown in Table 10. With the exception of Micrococcus citreus, all of the bacterial organisms showed some growth either in the broth or upon the agar. Both species of the molds showed good growth on both media, Aspergillus niger slightly more than Penicillium notatum. The three species of yeast and the five species of actinomycetes grew only slightly on these media.

Table 11 reveals the nutritive value of media prepared from Brassica oleracea extract. With the exception of Serratia marcescens and Neisseria catarrhalis, all species of bacterial organisms showed some growth on both of these media. Bacillus mycoides, Bacillus subtilis, Escherichia coli, and Aerobacter aerogenes grew the most luxuriantly of any bacterial organisms on the vegetable extract agar. The two species of molds grew on these media,

TABLE 10

NUTRITIVE VALUE OF BETA CRASSA
EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	++
Bacillus megatherium	+	++
Bacillus mycoides	+++	++
Bacillus subtilis	+++	+++
Gaffkya tetragena	+	-
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	++	+++
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	+
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	++	+
Escherichia coli	++	+++
Escherichia communior	-	+
Klebsiella pneumoniae	+++	+
Neisseria catarrhalis	+	+
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	++	+
Serratia marcescens	+++	+++
Aspergillus niger	+++	+++
Penicillium notatum	++	++
Saccharomyces cerevisiae	+	++
Saccharomyces ellipsoideus	-	+
Torula histolytica	++	+
Actinomycete X	++	+
Actinomycete 2	+	+
Actinomycete 4	++	+
Actinomycete 5	++	+
Actinomycete 44	+	+

Penicillium notatum more so than Aspergillus niger. Likewise all five of the species of actinomycetes grew on these Actinomycete 2 the most abundantly.

Table 12 shows the nutritive value of Brassica oleracea var. botrytis I extract culture media. On these Bacillus

TABLE 11

NUTRITIVE VALUE OF BRASSICA OLERACEA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	++	+
Bacillus megatherium	++	+
Bacillus mycoides	+++	+
Bacillus subtilis	+++	++
Gaffkya tetragena	+	+
Micrococcus pyogenes var. albus	++	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	++	+
Micrococcus citreus	+	+
Mycobacterium smegmatis	+	+
Aerobacter aerogenes	+++	+
Alkaligenes fecalis	++	+
Escherichia coli	+++	++
Escherichia communior	+	+
Klebsiella pneumoniae	+	+
Neisseria catarrhalis	-	+
Pseudomonas aeruginosa	++	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	++	+
Serratia marcescens	+	-
Aspergillus niger	+	+++
Penicillium notatum	+++	+
Saccharomyces cerevisiae	+	+
Saccharomyces ellipsoideus	-	++
Torula histolytica	-	+
Actinomycete X	-	+
Actinomycete 2	+++	+
Actinomycete 4	+	+
Actinomycete 5	++	+
Actinomycete 44	++	+

subtilis, Aerobacter aerogenes, Klebsiella pneumoniae, Pseudomonas fluorescens, and Pseudomonas aeruginosa grew profusely. Other bacterial organisms exhibiting some growth were Micrococcus pyogenes var. aureus, Micrococcus roseus, Escherichia coli, and Serratia marcescens. All of

TABLE 12

NUTRITIVE VALUE OF BRASSICA OLERACEA VAR. BOTRYTIS I EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	+
Micrococcus pyogenes var. albus	+	++
Micrococcus pyogenes var. aureus	++	++
Micrococcus roseus	++	++
Micrococcus citreus	-	+
Mycobacterium smegmatis	+	++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	++	++
Escherichia communior	+	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	+	+
Serratia marcescens	++	++
Aspergillus niger	+++	+++
Penicillium notatum	-	++
Saccharomyces cerevisiae	++	++
Saccharomyces ellipsoideus	+	++
Torula histolytica	+	++
Actinomycete X	++	++
Actinomycete 2	+++	+
Actinomycete 4	+	+
Actinomycete 5	+++	++
Actinomycete 44	+	++

the species of yeasts and molds showed light to medium growth; however, Aspergillus niger grew profusely. All of the five species of actinomycetes grew on both of these media, Actinomycete 5 and Actinomycete 2 the most abundantly.

According to Table 13 most of the test bacterial species grew moderately to heavily on the culture media prepared

TABLE 13

NUTRITIVE VALUE OF BRASSICA OLERACEA VAR. BOTRYTIS II EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	++
Bacillus megatherium	+	+++
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	+++	+
Micrococcus pyogenes var. albus	++	-
Micrococcus pyogenes var. aureus	++	-
Micrococcus roseus	++	-
Micrococcus citreus	++	+
Mycobacterium smegmatis	++	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	+++	+++
Escherichia coli	++	-
Escherichia communior	++	-
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	+
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	++	+
Serratia marcescens	++	++
Aspergillus niger	+++	+++
Penicillium notatum	-	+++
Saccharomyces cerevisiae	+++	-
Saccharomyces ellipsoideus	++	-
Torula histolytica	-	+++
Actinomycete X	+	+++
Actinomycete 2	++	-
Actinomycete 4	+++	+
Actinomycete 5	+++	+++
Actinomycete 44	+++	+++

from the extract of Brassica oleracea var. botrytis II. However, two bacterial organisms did not produce growth on the extract agar and five did not produce growth in the extract broth. Aspergillus niger, Actinomycete 5, and Actinomycete 44 grew luxuriantly on both these media.

Saccharomyces cerevisiae and Saccharomyces ellipsoideus exhibited good growth on the extract agar.

The nutritive value of Brassica rapa extract is shown in Table 14. Bacillus cereus, Neisseria catarrhalis, and

TABLE 14

NUTRITIVE VALUE OF BRASSICA RAPA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	+	+++
Bacillus mycoides	++	-
Bacillus subtilis	+++	+++
Gaffkya tetragena	++	+
Micrococcus pyogenes var. albus	+	-
Micrococcus pyogenes var. aureus	+	+
Micrococcus roseus	+	+
Micrococcus citreus	+	-
Mycobacterium smegmatis	-	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	+	+
Escherichia coli	++	+
Escherichia communior	+	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	+	+
Serratia marcescens	+++	++
Aspergillus niger	+++	+++
Penicillium notatum	+	-
Saccharomyces cerevisiae	++	+
Saccharomyces ellipsoideus	++	+
Torula histolytica	-	-
Actinomycete X	+	+
Actinomycete 2	+	+
Actinomycete 4	+	+
Actinomycete 5	++	+
Actinomycete 44	++	+

Torula histolytica showed no signs of growth on either one of the media prepared from this extract. Most of the other organisms grew only moderately. However, Bacillus subtilis, Aerobacter aerogenes, Klebsiella pneumoniae, and Aspergillus niger grew profusely on both types of media.

According to Table 15, which exhibits the nutrient value of Capsicum annuum extract media, abundant growth was

TABLE 15

NUTRITIVE VALUE OF CAPSICUM ANNUUM EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	+	-
Bacillus mycoides	-	+
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+	-
Micrococcus pyogenes var. aureus	+	-
Micrococcus roseus	+	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	-
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	+++	-
Escherichia communior	++	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	+	-
Serratia marcescens	+++	++
Aspergillus niger	+++	+++
Penicillium notatum	+++	+++
Saccharomyces cerevisiae	+++	+++
Saccharomyces ellipsoideus	+++	+
Torula histolytica	+	-
Actinomycete X	+	-
Actinomycete 2	+	-
Actinomycete 4	-	-
Actinomycete 5	+	+
Actinomycete 44	+	++

produced on these by Bacillus subtilis, Aerobacter aerogenes, Pseudomonas aeruginosa, Klebsiella pneumoniae, Pseudomonas fluorescens, and Serratia marcescens. Several of the species of bacteria as well as the strains of actinomycetes showed only poor to medium growth. Both species of the molds and the yeast, Saccharomyces cerevisiae, showed excellent growth on both media.

The nutritive value of Cucumis sativus extract is revealed in Table 16. On media prepared from this extract Bacillus subtilis and Aerobacter aerogenes showed medium to heavy growth, while several other species of bacterial organisms exhibited poor to medium growth. Six bacterial species did not grow at all on either medium. On the same media Aspergillus niger grew abundantly, while Penicillium notatum, the three species of yeasts, and the actinomycetes grew only slightly.

According to Table 17, which shows the nutritive value of the Cucurbita maxima extract, every test organism showed some growth either in the vegetable extract broth or on the vegetable extract agar. Bacillus subtilis, Aerobacter aerogenes, and Aspergillus niger grew abundantly in both types of media. All species of the actinomycetes showed medium to heavy growth in the extract media.

An observation of Table 18, which shows the nutritive value of Daucus carota extract, reveals that Bacillus

TABLE 16

NUTRITIVE VALUE OF CUCUMIS SATIVUS EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	+++	+
Gaffkya tetragena	-	+
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	++	+
Micrococcus citreus	-	+
Mycobacterium smegmatis	-	+
Aerobacter aerogenes	+++	++
Alkaligenes fecalis	-	-
Escherichia coli	++	+
Escherichia communior	-	+
Klebsiella pneumoniae	+	+
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	++	+
Pseudomonas fluorescens	+	+
Proteus vulgaris	-	-
Serratia marcescens	++	+
Aspergillus niger	+++	+++
Penicillium notatum	+	+
Saccharomyces cerevisiae	++	+
Saccharomyces ellipsoideus	++	+
Torula histolytica	-	+
Actinomycete X	+	+
Actinomycete 2	++	+
Actinomycete 4	+	+
Actinomycete 5	-	+
Actinomycete 44	+	+

subtilis, Bacillus megatherium, Bacillus cereus, Aerobacter aerogenes, and Escherichia coli grew extremely well on both media. Saccharomyces cerevisiae and Saccharomyces ellipsoideus showed no growth on the extract agar but exhibited

TABLE 17

NUTRITIVE VALUE OF CUCURBITA MAXIMA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	++	+++
Bacillus megatherium	++	+++
Bacillus mycoides	+++	+
Bacillus subtilis	+++	+++
Gaffkya tetragena	+	+
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	-	+
Micrococcus roseus	++	+
Micrococcus citreus	+	+
Mycobacterium smegmatis	+	+
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	++	+
Escherichia coli	++	+
Escherichia communior	-	+
Klebsiella pneumoniae	-	+
Neisseria catarrhalis	++	+
Pseudomonas aeruginosa	++	++
Pseudomonas fluorescens	-	+
Proteus vulgaris	++	+
Serratia marcescens	++	++
Aspergillus niger	+++	+++
Penicillium notatum	+	+
Saccharomyces cerevisiae	-	+++
Saccharomyces ellipsoideus	-	+++
Torula histolytica	++	+
Actinomycete X	++	++
Actinomycete 2	+++	+
Actinomycete 4	++	++
Actinomycete 5	++	++
Actinomycete 44	++	+++

abundant growth on the extract broth. All species of actinomycetes grew abundantly.

Nutritive value for the thirty test microorganisms on culture media prepared from the extract of Hibiscus

TABLE 18

NUTRITIVE VALUE OF DAUCUS CAROTA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	++	+++
Bacillus megatherium	++	+++
Bacillus mycoides	+++	+
Bacillus subtilis	+++	++
Gaffkya tetragena	+	+
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	+	+
Micrococcus roseus	++	+
Micrococcus citreus	+++	+
Mycobacterium smegmatis	++	+
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	++	+
Escherichia coli	++	+++
Escherichia communior	++	++
Klebsiella pneumoniae	-	++
Neisseria catarrhalis	+	+
Pseudomonas aeruginosa	++	++
Pseudomonas fluorescens	+	+
Proteus vulgaris	++	+
Serratia marcescens	++	++
Aspergillus niger	+++	+
Penicillium notatum	+	++
Saccharomyces cerevisiae	-	+++
Saccharomyces ellipsoideus	-	+++
Torula histolytica	++	+++
Actinomycete X	++	++
Actinomycete 2	+++	+++
Actinomycete 4	++	++
Actinomycete 5	++	++
Actinomycete 44	++	+++

esculentus is shown in Table 19. Two thirds of the test organisms grew moderately to luxuriantly on these media. Neisseria catarrhalis and Proteus vulgaris grew very sparsely. The two species of molds showed moderate growth,

TABLE 19

NUTRITIVE VALUE OF HIBISCUS ESCULENTUS EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	+++
Micrococcus roseus	-	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	+	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	+++	+
Escherichia communior	++	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	+	+
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	+	-
Serratia marcescens	+++	++
Aspergillus niger	++	+++
Penicillium notatum	+	+
Saccharomyces cerevisiae	++	++
Saccharomyces ellipsoideus	++	++
Torula histolytica	-	-
Actinomycete X	+	+++
Actinomycete 2	-	-
Actinomycete 4	-	+
Actinomycete 5	+	+
Actinomycete 44	+	+

Aspergillus niger the greater of the two. The two species of yeast, Saccharomyces cerevisiae and Saccharomyces ellipsoideus, showed medium growth. Actinomycete X, Actinomycete 5, and Actinomycete 44 exhibited only slight growth, but far surpassed that of the other actinomycetes.

The nutritive value of the extract of Lactuca sativa is shown in Table 20. None of the species of the genus

TABLE 20

NUTRITIVE VALUE OF LACTUCA SATIVA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	+
Bacillus megatherium	++	-
Bacillus mycoides	+++	+++
Bacillus subtilis	++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	-
Micrococcus roseus	-	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	+	++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	++	+
Escherichia coli	+	+++
Escherichia communior	+	++
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	++	++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	+	+
Serratia marcescens	++	++
Aspergillus niger	+++	+++
Penicillium notatum	+++	-
Saccharomyces cerevisiae	++	+
Saccharomyces ellipsoideus	++	+
Torula histolytica	-	-
Actinomycete X	+	+++
Actinomycete 2	++	-
Actinomycete 4	-	-
Actinomycete 5	++	++
Actinomycete 44	+++	+++

Micrococcus showed any growth at all. Bacillus mycoides, Bacillus subtilis, Aerobacter aerogenes, Klebsiella

pneumoniae, and Pseudomonas fluorescens exhibited heavy growth. Other organisms showing poor to medium growth were Mycobacterium smegmatis, Alkaligenes fecalis, Pseudomonas aeruginosa, and Serratia marcescens. Aspergillus niger grew profusely in both the extract broth and upon the agar; whereas Penicillium notatum gave a heavy growth only on the extract agar. The two species of Saccharomyces exhibited light to medium growth. Actinomycete 44 grew abundantly on both media.

Table 21 shows the nutritive value of the vegetable extract media of Lycopersicum esculentum. None of the gram-positive bacterial organisms nor the actinomycetes produced any growth either in the extract broth or upon the extract agar. Two gram-negative organisms, Aerobacter aerogenes and Klebsiella pneumoniae, grew luxuriantly on the extract media. Aspergillus niger and Penicillium notatum produced abundant growth, while Saccharomyces cerevisiae and Saccharomyces ellipsoideus exhibited only poor to medium growth.

The nutritive value of the extract media of Petroselinum hortense is revealed in Table 22. The test organisms grew extremely well on these extract media. Every organism exhibited growth in the extract broth; however, three organisms failed to grow on the extract agar.

TABLE 21

NUTRITIVE VALUE OF *LYCOPERSICUM ESCULENTUM* EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	-	-
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	-
Micrococcus roseus	-	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	-
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	+	++
Escherichia communior	+	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+	+++
Pseudomonas fluorescens	-	+++
Proteus vulgaris	-	-
Serratia marcescens	+	++
Aspergillus niger	+++	+++
Penicillium notatum	++	++
Saccharomyces cerevisiae	+	++
Saccharomyces ellipsoideus	+	++
Torula histolytica	-	-
Actinomycete X	-	-
Actinomycete 2	-	-
Actinomycete 4	-	-
Actinomycete 5	-	-
Actinomycete 44	-	-

Table 23 shows the nutritive value of *Phaseolus lunatus* extract media. With the exception of *Gaffkya tetragena* and *Micrococcus citreus* all of the test organisms produced growth on these media. Most of the organisms

TABLE 22

NUTRITIVE VALUE OF PETROSELINUM HORTENSE EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	+++
Bacillus megatherium	++	+++
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	+++	+++
Micrococcus pyogenes var. albus	+	+++
Micrococcus pyogenes var. aureus	++	+++
Micrococcus roseus	+	+++
Micrococcus citreus	+	+++
Mycobacterium smegmatis	-	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	+++	+++
Escherichia coli	+++	+++
Escherichia communior	+++	++
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	++	+++
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	++	+++
Serratia marcescens	+++	+++
Aspergillus niger	++	+++
Penicillium notatum	+	+++
Saccharomyces cerevisiae	+	+++
Saccharomyces ellipsoideus	+	+++
Torula histolytica	-	+++
Actinomycete X	++	+++
Actinomycete 2	+	+
Actinomycete 4	++	+++
Actinomycete 5	+	+++
Actinomycete 44	++	+++

revealed moderate to heavy growth; however, some organisms such as Micrococcus pyogenes var. albus, Micrococcus pyogenes var. aureus, Neisseria catarrhalis, and Proteus vulgaris showed very scant growth. Aspergillus niger and Torula histolytica grew luxuriantly both in the vegetable

TABLE 23

NUTRITIVE VALUE OF PHASEOLUS LUNATUS EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	+++
Bacillus megatherium	+++	+++
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	+	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	+	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	++	+++
Escherichia coli	++	+++
Escherichia communior	+++	+++
Klebsiella pneumoniae	++	+++
Neisseria catarrhalis	+	+
Pseudomonas aeruginosa	+	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	+	+
Serratia marcescens	++	+++
Aspergillus niger	+++	+++
Penicillium notatum	-	+
Saccharomyces cerevisiae	+	++
Saccharomyces ellipsoideus	+	++
Torula histolytica	+++	+++
Actinomycete X	+	+++
Actinomycete 2	+	-
Actinomycete 4	-	+
Actinomycete 5	++	++
Actinomycete 44	++	+++

extract broth and upon the vegetable extract agar. Actinomycete 5 and Actinomycete 44 showed more growth than any of this group.

The nutritive value of the extract media of Phaseolus vulgaris is shown in Table 24. Only five organisms failed

TABLE 24

NUTRITIVE VALUE OF PHASEOLUS VULGARIS EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	+++
Bacillus megatherium	+++	++
Bacillus mycoides	+++	+
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	++	+
Micrococcus citreus	-	-
Mycobacterium smegmatis	+++	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	+++	+
Escherichia communior	-	+
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	-	-
Serratia marcescens	++	++
Aspergillus niger	+++	+++
Penicillium notatum	+++	++
Saccharomyces cerevisiae	+++	++
Saccharomyces ellipsoideus	+++	++
Torula histolytica	+	+
Actinomycete X	+++	+++
Actinomycete 2	++	++
Actinomycete 4	+++	++
Actinomycete 5	+++	+++
Actinomycete 44	+++	+++

to grow in these media. Of the bacterial organisms, Bacillus megatherium, Bacillus subtilis, Mycobacterium smegmatis, Aerobacter aerogenes, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Pseudomonas fluorescens showed heavy growth. The two species of molds, Aspergillus niger

and Penicillium notatum, and two of the yeasts, Saccharomyces cerevisiae and Saccharomyces ellipsoideus grew profusely. All five species of actinomycetes grew luxuriantly both in the extract broth and upon the extract agar.

According to Table 25 approximately one half of the test organisms produced growth on the extract media of

TABLE 25

NUTRITIVE VALUE OF PISUM SATIVUM EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	++	+
Bacillus mycoides	+++	+
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+++	+
Micrococcus pyogenes var. aureus	+++	+
Micrococcus roseus	+	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	+	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	+++	+++
Escherichia communior	-	-
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	-	-
Pseudomonas fluorescens	+	-
Proteus vulgaris	-	-
Serratia marcescens	+	-
Aspergillus niger	+++	+++
Penicillium notatum	-	+
Saccharomyces cerevisiae	+++	++
Saccharomyces ellipsoideus	+++	++
Torula histolytica	+++	+++
Actinomycete X	+	-
Actinomycete 2	-	-
Actinomycete 4	-	-
Actinomycete 5	-	-
Actinomycete 44	-	-

Pisum sativum. Nine bacterial organisms exhibited medium to heavy growth on either or both of the extract media. The growth of Aspergillus niger was excellent on both types of media; however, Penicillium notatum showed only poor growth in the extract broth. The three species of yeast grew extremely well, but negative results were recorded for the species of actinomycetes.

An observation of Table 26 which shows the nutrient value of the extract media of Rheum rhaponticum reveals that the growth of the bacterial organisms was completely inhibited on both of the extract media. Aspergillus niger showed a heavy growth on the vegetable extract agar. Saccharomyces cerevisiae and Saccharomyces ellipsoideus revealed only slight growth on the agar; whereas Actinomycete X and Actinomycete 2 exhibited only barely perceptible growth.

Table 27 reveals the nutritive value of the extract media of Solanum melongena. Of the bacterial organisms, Bacillus mycoides, Bacillus subtilis, Aerobacter aerogenes, Klebsiella pneumoniae, Pseudomonas aeruginosa, and Pseudomonas fluorescens grew abundantly, while Gaffkya tetragena, Micrococcus citreus, Alkaligenes fecalis, and Neisseria catarrhalis showed no signs of growth. Aspergillus niger revealed excellent growth. Favorable results on the extract agar of the two species of yeasts, Saccharomyces

TABLE 26

NUTRITIVE VALUE OF RHEUM RHAPONTICUM EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	-	-
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	-
Micrococcus roseus	-	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	-
Aerobacter aerogenes	-	-
Alkaligenes fecalis	-	-
Escherichia coli	-	-
Escherichia communior	-	-
Klebsiella pneumoniae	-	-
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	-	-
Pseudomonas fluorescens	-	-
Proteus vulgaris	-	-
Serratis marcescens	-	-
Aspergillus niger	+++	+
Penicillium notatum	-	-
Saccharomyces cerevisiae	+	-
Saccharomyces ellipsoideus	+	-
Torula histolytica	-	-
Actinomycete X	+	+
Actinomycete 2	-	+
Actinomycete 4	-	-
Actinomycete 5	-	-
Actinomycete 44	-	-

cerevisiae and Saccharomyces ellipsoideus, was observed. Actinomycete 44 was the only one of the actinomycete group that gave any noticeable growth.

TABLE 27

NUTRITIVE VALUE OF *SOLANUM MELONGENA* EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	+
Bacillus megatherium	+	+
Bacillus mycoides	++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+	-
Micrococcus pyogenes var. aureus	+	-
Micrococcus roseus	+	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	++	-
Escherichia communior	++	-
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	+++	+++
Proteus vulgaris	+	-
Serratia marcescens	++	++
Aspergillus niger	+++	+++
Penicillium notatum	++	+
Saccharomyces cerevisiae	+++	+
Saccharomyces ellipsoideus	+++	+
Torula histolytica	-	-
Actinomycete X	+	+
Actinomycete 2	+	-
Actinomycete 4	+	-
Actinomycete 5	+	-
Actinomycete 44	++	+++

According to Table 28 which shows the nutritive value of *Spinacia oleracea*, *Penicillium notatum* exhibited poor growth. Both species of *Saccharomyces* grew moderately. In the actinomycete group only Actinomycete 44 showed any appreciable results on the extract media.

TABLE 28

NUTRITIVE VALUE OF SPINACIA OLERACEA EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	-
Bacillus megatherium	++	+
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	+	-
Micrococcus roseus	+	++
Micrococcus citreus	-	-
Mycobacterium smegmatis	+	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	++	+
Escherichia communior	++	+
Klebsiella pneumoniae	++	+
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	+	-
Serratia marcescens	++	+++
Aspergillus niger	+++	+++
Penicillium notatum	+	+
Saccharomyces cerevisiae	++	++
Saccharomyces ellipsoideus	++	++
Torula histolytica	+	+
Actinomycete X	-	-
Actinomycete 2	-	-
Actinomycete 4	+	-
Actinomycete 5	-	-
Actinomycete 44	+	++

Table 29 shows the nutritive value of Vigna sinensis extract media. Exceptionally poor growth was observed on these. Only nine organisms showed any signs of growth. The only significant one was Aspergillus niger. It grew abundantly.

TABLE 29

NUTRITIVE VALUE OF VIGNA SINENSIS EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	+	-
Bacillus mycoides	-	-
Bacillus subtilis	+	-
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	-
Micrococcus roseus	++	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	-
Aerobacter aerogenes	-	-
Alkaligenes fecalis	-	-
Escherichia coli	-	-
Escherichia communior	+	-
Klebsiella pneumoniae	-	-
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	-	-
Pseudomonas fluorescens	-	-
Proteus vulgaris	-	-
Serratia marcescens	-	-
Aspergillus niger	+++	+++
Penicillium notatum	-	-
Saccharomyces cerevisiae	+	-
Saccharomyces ellipsoideus	+	+
Torula histolytica	-	-
Actinomycete X	-	-
Actinomycete 2	-	-
Actinomycete 4	+	-
Actinomycete 5	-	-
Actinomycete 44	+	-

Table 30 reveals the nutritive value of the vegetable extract media of Zea saccharata. Each of the twenty bacterial organisms showed good growth with Bacillus mycoides, Bacillus subtilis, Aerobacter aerogenes,

TABLE 30

NUTRITIVE VALUE OF *ZEA SACCHARATA* EXTRACT FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	++	+
Bacillus megatherium	++	++
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	+++	+
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	+++	-
Micrococcus citreus	+++	-
Mycobacterium smegmatis	+++	-
Aerobacter aerogenes	+++	++
Alkaligenes fecalis	++	-
Escherichia coli	++	+
Escherichia communior	++	-
Klebsiella pneumoniae	+++	+
Neisseria catarrhalis	++	-
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	++	+++
Proteus vulgaris	++	-
Serratia marcescens	++	+
Aspergillus niger	+++	+++
Penicillium notatum	-	-
Saccharomyces cerevisiae	++	+
Saccharomyces ellipsoideus	++	+
Torula histolytica	-	+++
Actinomycete X	+++	+++
Actinomycete 2	++	+++
Actinomycete 4	++	+++
Actinomycete 5	+++	+++
Actinomycete 44	+++	+++

Pseudomonas aeruginosa, and *Pseudomonas fluorescens* exhibiting the heaviest growth on both types of media. Better growth was observed on the extract agar than in the extract broth. *Aspergillus niger* showed a very heavy growth; whereas *Penicillium notatum* did not grow on either the extract agar

or in the extract broth. Each of the five actinomycetes grew profusely.

An observation of Table 31 which shows the nutritive value of Mixture Number 1 reveals that Bacillus subtilis,

TABLE 31

NUTRITIVE VALUE OF MIXTURE NUMBER 1
EXTRACTS FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	+
Bacillus megatherium	+	+
Bacillus mycoides	+	+
Bacillus subtilis	+++	++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	++	+
Micrococcus citreus	-	-
Mycobacterium smegmatis	+	+
Aerobacter aerogenes	+++	++
Alkaligenes fecalis	+	+
Escherichia coli	++	++
Escherichia communior	-	-
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+++	+++
Pseudomonas fluorescens	++	++
Proteus vulgaris	+	+
Serratia marcescens	+++	+++
Aspergillus niger	+++	+++
Penicillium notatum	-	-
Saccharomyces cerevisiae	++	++
Saccharomyces ellipsoideus	++	++
Torula histolytica	++	++
Actinomycete X	-	-
Actinomycete 2	+	+
Actinomycete 4	+	+
Actinomycete 5	+	-
Actinomycete 44	+	+

Aerobacter aerogenes, Klebsiella pneumoniae, Pseudomonas

aeruginosa, Serratia marcescens, and Aspergillus niger exhibited abundant growth. The three yeasts exhibited moderate results.

The nutritive value of the extract media of Mixture Number 2 is shown in Table 32. Only three organisms

TABLE 32

NUTRITIVE VALUE OF MIXTURE NUMBER 2 EXTRACTS FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	-	-
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	-
Micrococcus roseus	-	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	-
Aerobacter aerogenes	-	-
Alkaligenes fecalis	-	-
Escherichia coli	-	-
Escherichia communior	-	-
Klebsiella pneumoniae	-	-
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	-	-
Pseudomonas fluorescens	-	-
Proteus vulgaris	-	-
Serratia marcescens	-	-
Aspergillus niger	+++	+++
Penicillium notatum	-	-
Saccharomyces cerevisiae	+	+
Saccharomyces ellipsoideus	+	+
Torula histolytica	-	-
Actinomycete X	-	-
Actinomycete 2	-	-
Actinomycete 4	-	-
Actinomycete 5	-	-
Actinomycete 44	-	-

exhibited any growth. These were Aspergillus niger, Saccharomyces cerevisiae, and Saccharomyces ellipsoideus, the first of these growing most profusely.

According to Table 33 which shows the nutritive value of Mixture Number 3 extract media, ten bacterial organisms

TABLE 33

NUTRITIVE VALUE OF MIXTURE NUMBER 3 EXTRACTS FOR THIRTY MICROORGANISMS

Organisms	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	++	+
Micrococcus pyogenes var. aureus	+	+
Micrococcus roseus	++	+
Micrococcus citreus	-	-
Mycobacterium smegmatis	+++	+++
Aerobacter aerogenes	+++	+++
Alkaligenes fecalis	-	-
Escherichia coli	+++	+++
Escherichia communior	-	-
Klebsiella pneumoniae	+++	++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	+	+
Pseudomonas fluorescens	-	-
Proteus vulgaris	-	-
Serratia marcescens	+	++
Aspergillus niger	+++	+++
Penicillium notatum	++	++
Saccharomyces cerevisiae	+++	++
Saccharomyces ellipsoideus	+++	++
Torula histolytica	+	-
Actinomycete X	+++	++
Actinomycete 2	++	++
Actinomycete 4	+	+
Actinomycete 5	++	++
Actinomycete 44	+	++

showed no signs of growth. However, the other organisms exhibited favorable results.

Table 34 reveals the nutritive value of the extract media of Mixture Number 4. Three gram-positive organisms,

TABLE 34

NUTRITIVE VALUE OF MIXTURE NUMBER 4
EXTRACTS FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	-	-
Bacillus megatherium	-	-
Bacillus mycoides	-	-
Bacillus subtilis	+++	+++
Gaffkya tetragena	-	-
Micrococcus pyogenes var. albus	-	-
Micrococcus pyogenes var. aureus	-	-
Micrococcus roseus	+	-
Micrococcus citreus	-	-
Mycobacterium smegmatis	-	+
Aerobacter aerogenes	++	+++
Alkaligenes fecalis	-	-
Escherichia coli	-	-
Escherichia communior	-	-
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	-	-
Pseudomonas aeruginosa	-	-
Pseudomonas fluorescens	-	-
Proteus vulgaris	-	-
Serratia marcescens	-	-
Aspergillus niger	+++	+++
Penicillium notatum	++	++
Saccharomyces cerevisiae	+++	++
Saccharomyces ellipsoideus	++	+
Torula histolytica	-	-
Actinomycete X	-	-
Actinomycete 2	-	-
Actinomycete 4	+	+
Actinomycete 5	-	-
Actinomycete 44	-	-

Bacillus subtilis, Micrococcus roseus, and Mycobacterium smegmatis, exhibited growth with Bacillus subtilis showing the heaviest growth. Only two gram-negative organisms, Aerobacter aerogenes and Klebsiella pneumoniae, grew. Both Aspergillus niger and Penicillium notatum showed good growth.

TABLE 35

NUTRITIVE VALUE OF MIXTURE NUMBER 5
EXTRACTS FOR THIRTY MICROORGANISMS

Organism	Extract Agar	Extract Broth
Bacillus cereus	+	+
Bacillus megatherium	++	+
Bacillus mycoides	+++	+++
Bacillus subtilis	+++	+++
Gaffkya tetragena	+++	++
Micrococcus pyogenes var. albus	+	+
Micrococcus pyogenes var. aureus	++	+
Micrococcus roseus	++	++
Micrococcus citreus	+++	++
Mycobacterium smegmatis	+++	++
Aerobacter aerogenes	+++	++
Alkaligenes fecalis	+++	+++
Escherichia coli	+++	++
Escherichia communior	+++	++
Klebsiella pneumoniae	+++	+++
Neisseria catarrhalis	++	+
Pseudomonas aeruginosa	++	++
Pseudomonas fluorescens	++	++
Proteus vulgaris	++	+
Serratia marcescens	+	+
Aspergillus niger	+++	+++
Penicillium notatum	-	-
Saccharomyces cerevisiae	++	+
Saccharomyces ellipsoideus	+	-
Torula histolytica	-	-
Actinomycete X	+++	+++
Actinomycete 2	+++	+++
Actinomycete 4	+	-
Actinomycete 5	+++	+++
Actinomycete 44	+++	+++

The two species of Saccharomyces grew moderately; whereas in the species of actinomycetes only Actinomycete 4 showed any growth.

The nutritive value of the extract media of Mixture Number 5 is shown in Table 35. Every bacterial organism exhibited growth, the least being produced by Bacillus cereus, Micrococcus pyogenes var. albus, and Serratia marcescens. Aspergillus niger exhibited excellent growth, while Penicillium notatum and Torula histolytica gave negative results. Each of the actinomycetes, with the exception of Actinomycete 4, grew abundantly.

CHAPTER IV

SUMMARY AND CONCLUSIONS

Summary

1. Twenty-five different kinds of common garden vegetables were obtained and extracts of these were used in the preparation of laboratory culture media for use in this investigation.

2. Thirty species of representative microorganisms were used in testing for nutritional value in these vegetable extracts.

3. The test microorganisms were cultured on Tryptose agar slants and in Tryptose agar broth as stocks. Inoculations were made from the Tryptose broth on to the extract agar plates and from the Tryptose agar slants into the extract broths.

4. Mixtures of several of the vegetable extracts were also used in order to determine the nutritive value of the extracts when combined in one culture medium.

5. The pH of each vegetable extract was determined and recorded.

6. The results of growth, as criteria for determining nutritive values, were observed and evaluated after an

incubation period of forty-eight hours. These were recorded in tabular form.

7. Among the gram-positive bacterial species used in this investigation Bacillus mycoides and Bacillus subtilis showed consistently good growth on the greatest number of the vegetable extract media tested. Micrococcus citreus, Bacillus cereus, and Gaffkya tetragena grew consistently less vigorously than the other gram-positive species. Vegetable extract media prepared from Cucumis sativus, Capsicum annuum, Hibiscus esculentus, Vigna sinesis, Mixture Number 2, Mixture Number 4, Rheum rhaponticum, and Lycopersicum esculentum proved the least satisfactory in nutrients for the gram-positive organisms. The last two of these completely inhibited the entire group. The extract media prepared from Daucus carota, Petroselinum hortense, Zea saccharata proved the most valuable for the gram-positive group.

8. Gram-negative bacterial species did not grow as consistently well on the vegetable extract media as did the gram-positive ones. Alkaligenes fecalis, Neisseria catarrhalis, Escherichia communior, and Proteus vulgaris did not grow at all on many of the media tested. The extracts of Vigna sinensis, Cucumis sativus, Pisum sativum, Rheum rhaponticum, Mixture Number 2, and Mixture Number 4 revealed consistently poor nutritive content for the gram-negative species. The most valuable extract media for this

group of organisms were prepared from Allium cepa, Beta crassa, Brassica oleracea var. botrytis II and Petroselinum hortense.

9. Aspergillus niger grew abundantly on a majority of the extract media tested; Penicillium notatum grew somewhat less vigorously. Capsicum annuum and Phaseolus vulgaris contained nutrients which supported excellent growth for both of these species.

10. The growth of Saccharomyces cerevisiae and Saccharomyces ellipsoideus varied considerably on the various extract media. However, Phaseolus vulgaris and Pisum sativum extracts supported excellent growth for both of these species. Torula histolytica grew well on culture media prepared from extracts of Phaseolus lunatus, Daucus carota, and Pisum sativum.

11. The most significant growth exhibited by the strains of actinomycetes was noted on extract media prepared from Brassica oleraceae var. botrytis II, Cucurbita maxima, Daucus carota, Phaseolus vulgaris, Zea saccharata, and Mixture Number 5. The extract medium of Lycopersicum esculentum completely inhibited the growth of all the strains of actinomycetes.

Conclusions

From growth criteria determined by this investigation the extract media prepared from Daucus carota, Petroselinum

hortense, and Zea saccharata have been found the most valuable for the cultivation of the gram-positive species of bacteria. Those prepared from the extracts of Allium cepa, Beta crassa, Brassica oleracea var. botrytis II, and Petroselinum hortense have been found the most satisfactory for growing the gram-negative bacterial species. Whereas most all of the extract media were found satisfactory for the growth of Aspergillus niger and Penicillium notatum, those prepared from Capsicum annuum and Phaseolus vulgaris were found the most significant. Phaseolus vulgaris and Pisum sativum extract media supported excellent growth for both Saccharomyces cerevisiae and Saccharomyces ellipsoideus. Torula histolytica grew well on culture media prepared from extracts of Phaseolus lunatus, Daucus carota, and Pisum sativum. Extract media prepared from Brassica oleracea var. botrytis II, Cucurbita maxima, Daucus carota, Phaseolus vulgaris, Zea saccharata, and Mixture Number 5 were found highly satisfactory for culturing the strains of actinomycetes.

These results indicate that certain vegetable extracts can be used in the preparation of suitable and economical culture media which can be used both on the secondary and higher education level for the growth of microorganisms. Further investigations should be made in order to determine the amino acid, metallic ion, accessory

growth factor, and carbohydrate content of these vegetable extracts in order to determine whether these correspond to those, set down by others, as the nutritional requirements for microorganisms.

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