CHAPTER II

AGAR

2.1 Introduction

Agar was first discovered about 1658 when the Japanese Emperor was marooned at an inn during a snow storm (5). The innkeeper Tarozaemon Minoya prepared seaweed jelly for the Emperor's dinner and threw the excess outside on the snow and evidently froze during the night. The next morning the frozen jelly thawed and turned into a dry papery translucent substance, which the innkeeper found could be reconverted to its jellied form.

According to the U.S. Pharmacopeia (6), Agar is a hydrophilic colloid extracted from marine algae of the class Rhodophyceae. Agar is insoluble in cold water but soluble in boiling water. A 1.5% solution is clear, and when cooled to 32-39^{o}, it forms a firm, resilient gel that does not melt below 85^{o}. In the interest of explicitness, the term agar will be reserved here for polysaccharides so defined. Other gums resembling agar, but not meeting all the specification of this definition,will be refered to as agaroids.

Two things are commonly called by the name of agar-

agar. On the one hand it is the well-known name of a trade product in Europe, which is prepared from certain species of red algae, but on the other hand it is also used on occasion for drugs prepared from untreated red algae which have merely been dried. There is no doubt that much confusion exists in the use of the term and likewise there is also much erroneous information about its sources. The word is of Malayan origin and in that language it refers to red seaweeds of the genus Eucheuma. Hoffman states that the word agar in Malay referred to the red alga Gracilaria lichenoides, but this statement is not correct as Eucheuma is the genus most widely used in Malaya for making an agar-like material.

2.2 Source

The untreated algae are differentiated according to the country of origin (5), e.g. Ceylon agar or Ceylon moss. This affords an example of the use of the word agar as applied to untreated red algae, a usage which should be agandoned. Ceylon moss refers to the dried red seaweed Gracilaria lichenoides obtained primarily in the island, though it also grows on the coasts bordering the Indian Ocean, where it is called Bengal isinglass. Chinese moss refers to another species of Gracilaria, G.verrucosa. In Japan agar-agar, made

from Gelidium spp., goes under the name of 'Kanten', which means 'cold sky', and as such refers to the fact that the material used to be prepared on cold winter days, or else high up on the mountains where it is always cold. Macassar agar, Java agar, algal-algal, or East Indian carragheen, which is not a true agar, comes from the Malay orchipelago, where the principal source is the red seaweed Eucheuma muricatum f. depauperata (= E. spinosum or E. denticulatum in some of the literature) though other species of Eucheuma may also be employed. The various islands in the orchipelago use different names of the algae employed in the production of agar.

2.2.1 Raw material

Agar is obtained from various genera and species of the red-purple seaweeds, class Rhodophyceae, where it occurs as a structural carbohydrate in the cell walls and probably also performs a function in ion-exchange and dialysis processes.

i) Species

It will be realized, therefore, that agar is not derived from even a single algal genus, and it may be expected to be diverse in its characters and properties. Agarophytes of major commercial value are listed in Table 2-1 (5,6). It is very important, therefore, that in all investigations concerning this material, and in chemical analysis, the exact species and even variety should be clearly stated.

ii) Habitat

These seaweeds, in Table 2-1, are found from the intertidal zone to depths of more than 40m. Growth is most prolific in areas of surface turbulence and of marked top to bottom exchange. The plants grow from hold fasts attached to rocky substrates and attain lengths of from 0.1 to 2 m. Gelidiales usually shown an alternation of isomorphic generations, and propagation may be either by spores or stolons.

iii) Harvesting (Collections)

Harvesting from Mexican and some Japanese waters is done by divers in full pressure suits who tear the individual planets from their hold fasts and place them in rope bags in which they are raised to the surface (5). A diver is usually served by two helpers in a boat that is provided with an air compressor, emergency gear, weed storage space, and motive power. The diver must often work under the formidable handicaps of jugged to pography, strong currents, and low illumination. Despite these conditions, he must maintain proper buoyancy and select only suitable plants as he moves with or against a bottom current or wave surge. In this sort of operation, an expert diving term can gather from 200 to 1,000 kg at wet weed per 6-hr diving day. When dried, this output will range between 40 and 320 kg.

In Japan, the seaweeds are gathered from rocks between

Table 2-1 Rhodophytes used in the Agar industry

Species	Country	
Acanthopeltis japonica	Japan sea	Toriashi-agar
Ahnfeltia plicata	White Sea, Sakhalin	Base of Rusian industry
Gelidiella acuosa	Japan	Agar & food
Gelidium caulacanthum	New Zealand	
Gelidium coulteri	Mexico	
Gelidium corneum var sesquipedale	Spain, Portugal, Morocco, California	
G. amansii	Japan	Tengusa
G. divaricatum	"	
G. japonicum	"	Mixed with G. amansii as adulterants
G. liatulum	"	
G. pacificum	"	
G. subfastigiatum	"	
G. Vagum	"	
G. cartilagineum	South Africa, Mexico, California	Base of U.S.A. agar industry
G. medifrous	California	Used as adulterants
G. arbaresceus		
G. densum		
G. lingulatum	Chile	
G. pristoides	South Africa	
G. spinulosum	Morocco	
Gracilaria verrucosa (confervoides)	Atlantic, N.America	Principal species
	California, S. America, India, Ceylon, Japan, China, Formosa, Phillipines	Important in U.S.A. Japan & Australia
	Australia	

mid-and Low-tide marks, with long handled rakes in shallow waters, and divers collect them from the sublittaral regions. Women divers can collect down to a depth of 30 ft. with the use only of goggles. The best months for collection are July and August, though havesting actually takes place continuously from May to October. The best weed comes from the deeper waters. After the weed has been collected it is dried on the shore and is partly bleached, and then sold to the factories. The rights to gather agarophytes are controlled by the Central Federation of Fisherman's Co-operative Association and they also are responsible for distributing the weed to the various processing factories. Originally it was stored until the arrival of the cold winter months, but now a days with refrigeration this is not necessary.

In Baja California (6), America, harvesting is carried out by divers who collect the plant by hand. Each boat has a crew of three-diver, boat operator, and life-lime tender. The alga is put into a rope basket which is hauled to the surface when full (it then contains 60-70 lb of seaweed). In a single working day a diver can harvest up to 1½ tons of fresh Gelidium, but quantities much less than this are move normal. Diving can only be carried out when the water is calm and without a strong ground swell. Because of this most of the collecting is restricted to the period between May and November. Some companies furnish the boat and facilities and pay the

harvesters per fresh ton ($85 in 1945), whilst in other cases the harvesters provide the boat and equipment. In the latter case they also dry and bale the weed. Under these conditions the harvesters received from $350 to $400 per ton (1945) of sun-dried seaweed.

In Australia, a special grapnel has been devised for its collection, and this would appear to be the only case of a special travel designed to collect these smaller seaweeds. The basic structure is the Agassiz travel with a two-inch mesh net so that the sand can be washed out whilst collecting, a process also facilitated by traveling against the tide. A glass buoy is attached to the net as a direction indicator and also as a salvage line should the two ropes break. It has been found that the best results are obtained by using a motor-boat with a 7 h.p. engine with a towing rate of a quater speed. This type of equipment works satisfactorily in 6-100 ft. of water, which is probably as deep as one would require. In order to give some idea of its efficiency, it can be stated that hauls made in Botany Bay during the course of one hour produced 600 lb of wet weed, whilst in another bay three hauls, occupying half an hour in all, brought in a harvest of 400 lb. Up to 8 tons of wet weed can be collected in one week by a crew of three men. When the boats return to harbour, the weed in laid out on wire-netting racks to dry. Drying is important because extracts of

wet weed do not gel. Some times it is dried by artificial heat but in such cases the products is not so good.

In other parts of the world, a preponderence of weed is harvested by wonders at low tide, raked from rowboats, or picked by skin divers, who may wear either unventilated face masks or be supplied with compressed air from the surface in the manner of the full pressure suit divers. After the seaweed is gathered, it is freed of unwanted plants, stones, and other detritus; washed with fresh water when available; and spread in 1-5 cm. layers on sands, grass, or racks to dry. In average weather, with daily inversion of the layers, the moisture content will have dropped to less than 20% in 4 days and the weed will have been partially bleached by solar radiation. At this point, most weed is baled, although some may be washed and redried until thoroughly bleached before boiling. The baled weed, if the moisture content is maintained below 20%, will have a storage life of approximately 5 years.

iv) Price

The major commercial weeds, which may contain 30 to 45% agar, are frequently in short supply, which causes their market prices to undergo substantial fluctuations (6). The raw material used in the United States is perchased directly from foreign and domestic agents on the basis of analysis of representative samples taken at time of delivery. The prices

paid are based on the agar content, gel strength, and the percentage of contaminants and may vary from $500 to $1,250 per 1,000 kg delivered. The minor varieties, usually containing 15 to 30% agar, will follow these variations and will command from $350 to $700 per 1,000 kg.

2.2.2 Processing

Agar is insoluble in cold water but is colloidally dispersible in water above 90° (6). When agar gel are frozen, the agar skeleton contracts toward the center of mass as a membrane, leaving the ice as a separate phase. The ice will, in general, contain ten times more soluble salts, sugars, and simpler gums than is retained by the agar membrane. Hence, most commercial agar is manufactured by hot water extraction followed by freezing for purification. Other methods are possible, such as extraction with glycerol, anhydrous ammonia, or other solvents, and the use of alcohols and other flocculants to avoid the freezing operation.

In the traditional process, great care is exercised in blending the various algae chosen for a batch in an attempt to obtain a product of the desired flexibility, luster density, and surface smoothness and of the usual physical properties of gel solidity and resilience. Usually six or seven types of weed are used.

Operation begins soon after freezing nights are assured. The weeds are washed in batches of 8 kg and pounded for 20 min

to remove any sand and epiphytes. For each 200 kg of seaweed, about 2200 liters of water are boiled in an open iron caldron over a pine log fire. The tougher types of algae are introduced into the caldron first and the softest last. When the intermediate weeds have been added, the mixture is treated with 1 g of sulfuric acid or 0.3 g of polyphosphate per kg of seaweed to adjust the pH to 5-6 and extraction continues at 80° for 8-9 hr; at that time, weak liquor from the previous day is added. At approximately the twelfth hour, calcium hypochlorite or sodium bisulfite is introduced for bleaching at a rate of 2g per kg of weed, and the cooking is continued to the fifteenth hour.

The entire cook is strained through cloth of 3mm mesh and the cake is pressed and retained for recooking. The liquor containing 1% of agar is cleared somewhat by sedimentation, after which it is allowed to solidify in 170 × 30 × 1-cm wooden trays.

The gel is cut into strips, laid on straw mats outdoors and allowed to freeze, the strips being covered if the temperature is too low. Each day some of the night-formed ice melts, taking with it some salts, nitrogeneous material and residual color. Sprinkling is used when needed to prevent excessively rapid drying. After 5-6 days, the racks carrying the mats are orientated with respect to the sure in such a manner that final drying is complete in another 15-30 days.

In order to prepare an acceptable product, the manufacturer must be constantly alert to count the effects of climatic changes. Rapid freezing causes poor luster and feel. The surface ice must be broken by tapping to prevent the formation of sharp, fragile corners. Poor color results if either freezing or drying is too rapid.

The traditional method is essentially a cottage industry most installations being one or two cooker ventures and may being one-family enterprises. 005210

Since 1945, several firms throughout world have been a more scientific agar manufacture. Each manufacture generally uses various locally developed modifications of the basic extraction-freezing-thawing-drying method. In the main, the newer methods employ countercurrent and cascade multiple extraction, centrifugation, plate-and-frame press filtration, artificial freezing,chemical bleaching, drying with hot air by drum and spray methods, and grinding.

In the United states, the following sequence of operations is employed: (a) cleaning raw material, (b) chemical pretreatment, (c) pressure extraction, (d) chemical posttreatment, (e) filtration, (f) gelation, (g) freezing, (h) posttreatment, (i) washing, (j) drying, (k) sterilization, (l) bleaching, (m) washing, (n) drying.

2.2.3 Manufacture

At Shimizu-mura, Japan, a monument commemorates the first commercial manufacture of agar by a relative of Tarozaemon,

Miyata Hanbei of Aza Shiroyama. The Japanese industry expanded slowly until, in 1940, there were 400 processors. Manufacture of agar started in china and on the Malay Peninsula in 1850. Irregular production began in India, Ceylon and Australia in about 1880-1900, and in about 1915, Indonesian production was established.

i) Agar industry in Japan

The world production of agar must be currently about 5,500 metric tons annually, and of this Japan Produces about 3,500-3,600 metric tons. Half of this is manufactured industrially by some fifteen commercial firms, whilst the remainder is produced by about 400 small-scale operators. The principal centres of productions are in Hokkaido and at Osaka, Kyoto, Hyogo and Nagano.

When the manufacture was first commenced, agar was simply a mass of jelly obtained by boiling seaweed, but now it appears on the market in the form of sticks, sheets and bars, a manner of preparations which was taken up quite accidentally, due to the fact that one dry some jelly was put out of doors and solidity in these shapes.

After the weed has been collected it is dried on the shore and is partly bleached. Further treatment is as follows: it is first of all cleaved by beating and pounding, whilst larger lumps of foreign material are picked out by hand. Finally it was washed in running fresh water, after which it

is laid out to bleach on mats. This is done in warm weather, beginning in August, and the bleaching is much aided by dew If the conditions are very favourable, twenty-four hours may be sufficient for the process, but it usually takes several days. The more modern method is to wash and stir in the weed in vats, keeping the water temperature below 10°C. The 'hard' weeds (Gelidium spp.) require about 25 hr and the 'soft' weeds (e.g. Gracilaria) 10-15 hr. The importance of blanching for ordinary commercial use is exaggerated because it is not really necessary: it is, however, desirable for bacteriological purposes. As the drying and bleaching goes on the algae fuse into thick or thin sheets which can be loosely rolled. These sheets are then input into large wooden or iron vats together with some water, the exact quantity depending on the condition of the seaweed. At this stage blending of the weeds takes place.

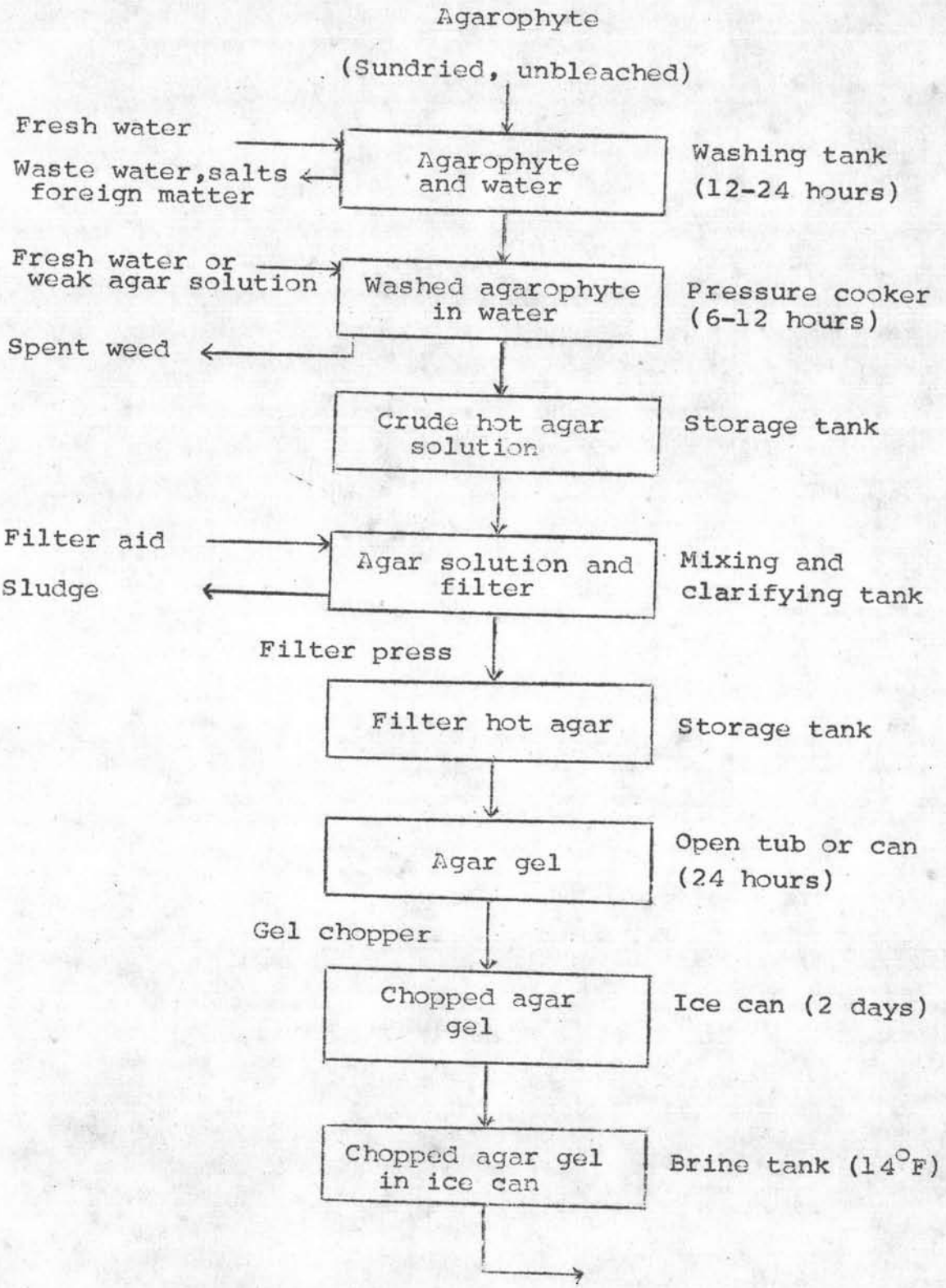
The final product is therefore not a pure kanten produced from a single species of Gelidium. Not only may a mixture of local algae be used but agarophytes imported from Indonesia are commonly added. The amount of adulteration varies from 20 to 40%, the exact quantity depending on the purpose for which the agar is required.

In extraction process, sulfuric acid is first added in order to control the pH since this affects the quality of the agar. The 'hard' weeds (Gelidium) are boiled for 1½hr

and then the 'soft' weeds added and boiled for 15 min, after which the whole mixture is allowed to simmer for 12hr. The liquor is strained first through a course cloth and then through a fine linen cloth under pressure, and the weed left is subjects to a second boiling of about 10hr. At the very end it is bleached by adding sodium peroxide. It is again filtered and the liquor is poured into wooden trays to cool by making use of rectangular wooden vessels. The cold jellied material is known as 'Tokoro-Ten'

A freezing temperature is now essential for the next stage, but before being subjected to the freezing process, it is cut up by means of oblong iron frame with sharpened edges into suitable sizes for further handling; in this state it is known as bar kanten or 'Kaku Kanten'. Some of this is exported to Holland where it is largely used in the manufacture of beer. When the bar kanten thaws after being frozen, the water flows away with the impurities, having behind relatively pure agar-agar, which is then dried. The frozen process, either in the open or in refrigerated rooms, takes from one to three days and another three or four days are allowed for drying.

The kaku kanten is also put into cylinders and pressed out through a perforated base into thin fine threads 30-35 cm long, looking rather like macaroni. This material is known as 'Huoso' or 'Hoso Kanten'. The kanten is finally parcelled


Agarophyte
(Sundried, unbleached)
Fresh water
Waste water, salts foreign matter
Agarophyte and water
Washing tank (12-24 hours)
Fresh water or weak agar solution
Spent weed
Washed agarophyte in water
Pressure cooker (6-12 hours)
Crude hot agar solution
Storage tank
Filter aid
Sludge
Agar solution and filter
Mixing and clarifying tank
Filter press
Filter hot agar
Storage tank
Agar gel
Open tub or can (24 hours)
Gel chopper
Chopped agar gel
Ice can (2 days)
Chopped agar gel in ice can
Brine tank (14°F)

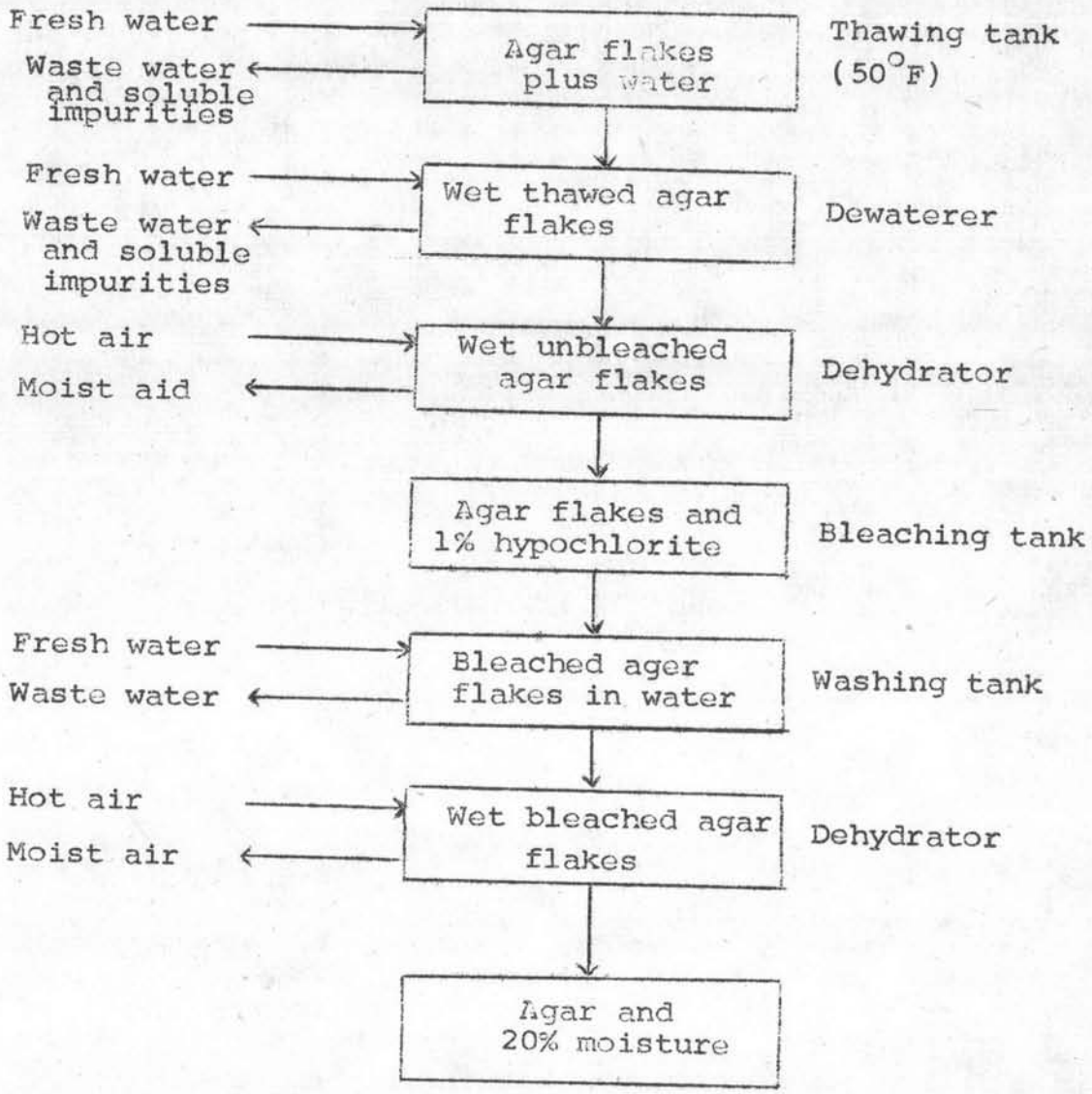


Figure 2-1 Flow diagram of the American process of agar manufacture as used in California. The bleaching portion is used by only one firm.

up and made into bales weighing 133lb, but it is sometimes sold in a shredded or powdered form. The commercial product is pearly white, shining and semi-transparent, and is also tasteless and odourless.

ii) Agar industry in America

The preparation of agar from seaweed in America follows much the same sequence as the modern Japaneses process. If the species of Gigartina are used, a preliminary soaking is first of all necessary followed by a boiling in a solution containing 2% chloride of lime. Unless this is carried out the resulting jelly is not firm enough for commercial purposes. This is because the product is not a true agar. Matsuoka, the man who laid the foundation of the now flourishing U.S. industry, not only introduced artificial freezing, but he also substituted bleaching by chlorine for sun bleaching. The value of this method of bleaching is open to some doubt because of possible effects upon the gel strength.

The Gelidium is first washed and soaks for 12-14 hr, after which it is transferred to special pressure cooker, where it is cooked for 6 hr at a pressure of 15 lb to the in^2 in a dilute agar solution from the third and final cook. It then receives two more cookings before being discarded. The extract is clarified and filtered, and then poured into open tanks where it gels after 24 hr. The gel is chopped up and

put in cans in a freezing rooms at 14°F for about two days. On removal it is thawed and placed in the dewaterer, which removes water with the soluble impurities.

The purified agar flakes, which now contain about 90% moisture, are dried by hot air until they contain about about 35% moisture. After this they are bleached in 1% sodium hypochlorite solution at room temperature. The excess bleaching reagent is reduced by sodium sulphite, after which the agar is removed, washed, and finally dried until only about 20% moisture remains.

In the original Becker process there was a special combined congealer and sizer, which was used to cool and gel the solution. This has now been abandoned in favour of the older method of cooling and gelling in open tubs. Also in the original process activated carbon was used to decolorize the agar. Unless great care was taken, particles of carbon passed into final product, and so the function of the carbon has been replaced by the bleaching process.

Work on agar seaweeds (7) has shown that pH of the extracting solution is of vary considerable important, but the effect varies for the different species. Thus the yield from Gelidium cartilagineum and P. terocladia species is maximal around pH 6-8 and falls off with increasing alkalinity. In the case of Endocladia muricata the reverse holds true, the maximum yield being secured at pH 12. Attempts to extract and agar-like substance from Gigartina canaliculata and G. senata were only successful when they adopted the treatment used by

Kizevetter (1937) to secure agar from Ahufeltia plicata on the maritime coast of Russia and from Phyllophara rubens in the Black sea.

On the east coast of America an agar industry using Gracilaria had made rapid headway. In the first nine months in 1944, 25,000 lb were produced. Extraction must take place under steam pressure or by boiling, and it would seem that although the extraction is easier than with Gelidium the amount extracted is not so great. Not only does the agar content vary with the season, but so must the methods of extraction.

iii) Agar industry in Australia

In the last century a company was formed at Dongarra in Western Australia for the purpose of making agar from the red seaweed Eucleuma speciosum.

Extraction is done by the weed is treated by boiling with steam in open vats, with mechanical stirrers in order to avoid 'cold spots', and great care has to be taken to control the acidity of the solution since below pH 5 the yield falls off rapidly. It has also been found that the use of iron and copper vessels tends to make the agar discoloured. The acidity of the solution is controlled by using sodium acid phosphate and an extraction of 2-4 hr. After filtering, re-digesting and re-filtering the liquor is clarified by activated carbon. It is then subjected to freezing, thawing and

dewatering. The most satisfactory drying process, evolved by the Karna Vita Company, is to wash in alcohol and dry under infra-red lamps. It does not provide such a good agar as the Japanese product but for meat canning it is regarded as superior. The arsenic content is low and it can be used for most bacteriological work. Bleaching does not seen to produce a better product and in fact it reduces the yield. It requires 7 tons of wet drained Gracilaria to produce one ton of dry dark weed, but 8 tons of wet weed are necessary in order to produce one ton of bleached weed. One ton of agar can then be produced by boiling 3 tons of the dark dried seaweed (therefore 21 tons of wet weed yield one ton of agar), and the product is said to be not inferior to imported Japanese agar. Some idea of the labour involved can be gauged from the fact that it is estimated that twenty men can harvest, dry and bleach 8 tons of wet weed per day. Generally speaking, the economics of Australian agar do not enable it to compete very well with imported Japanese agar.

2.3 Chemistry of agar

The structure of agar has been under study for decades, primarily by Japanese chemists (8). Whistler and Smart (9) stated in 1953:

The complete structure of agar is not established.

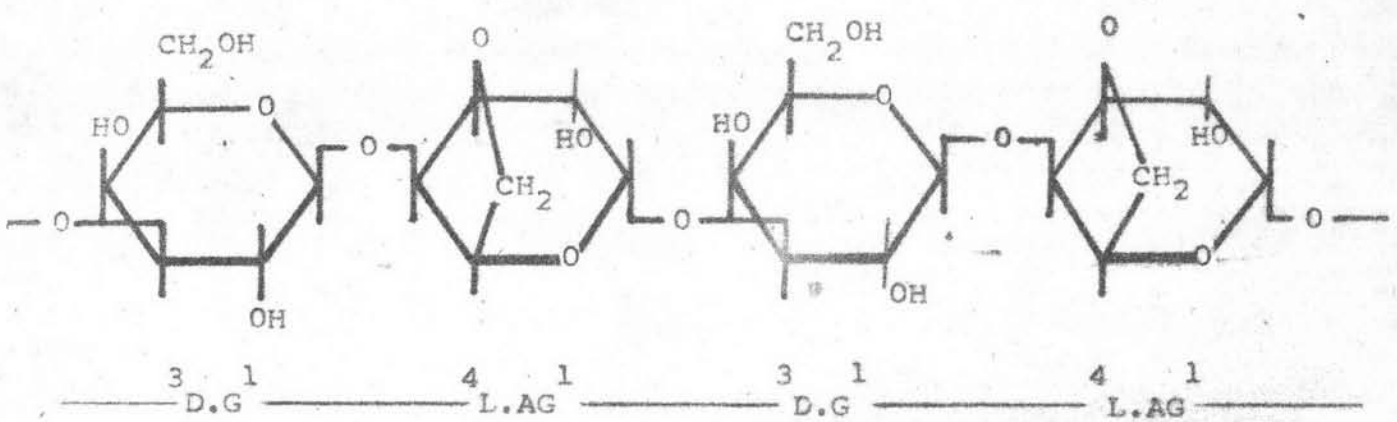
However, as late as 1970, Percival states that the structure of agar was unresolved which points up the difficulties in the structural elucidation of agal polysaccharides in general (10). Much of the difficulty lies in the extraction procedures where, due to the similarities in structure, a polysaccharide thought to be pure may in reality be a mixture. There is also the possibility of altering the structure of the polysaccharide during extraction of hydrolysis preceding analysis. Those polysaccharides containing sulfate ester are particularly susceptible to desulfonation and alteration of structure (11).

Acetylation and fractionating procedures have shown the presence of at least two components in the agar-extract. One of these, agarose, is a neutral polymer consisting of repeating units of β-D-galactopyranose residues connected through 1-and 3-positions and 3,6-anhydro-α-L-galactopyranose residues connected through 1-and 4-positions shown in Figure 2-2 (12). The other fraction, agaropectin, possibly a mixture of polysaccharides, comprises mainly D-galactose, 3,6-anhydro-L-galactose, some ester sulfate (3.5 to 9.7%) and D-glucoronnic acid(13). Recent studies have indicated that it may well be a complex mixture (14,15).

2.4 Properties

2.4.1 Solid

Solubility -At 25°, agar of high purity is practically insoluble in water. It is very slightly soluble in ethanolamine



D.G. = β-D-Galactopyranose

L.AG = 3,6-Anhydro-α-L-galactopyranose.

Figure 2-2 Agarose

and soluble in formamide

If agar, agarose, or Gracilaria agaroid is flocculated from a warm dispersion by five to ten volumes of ethanol and drained but not allowed to dry, it is soluble in water at 25° and will form a gel without the need for heating Agar so flocculated is also soluble at 25° in other solvents, but gelation will not occur until water is added. Dry agar is colloidally dispersible in water and in other liquids at 97-100°, and moist agar flocculated by ethanol, 2-propanol or acetone, or salted out by high concentrations of electrolytes, is soluble in a number of liquids.

Dispersions of 0-5% are conveniently made by heating on a steam or boiling water bath with occasional agitation, on a hot plate or over a low flame with constant stirring, by stagnant auto claving at 100°- 120° or, best, by the use of spherical containers with sigmental agitators heated by electric mantles or steam jackets.

For the preparation of heavier dispersions, such as 8-14% monlage mixtures, a vertical, jacketed, metal cylinder, provided with packing glands at both ends and a perforated piston agitator mounted in the center of a piston rod passing through both glands, is convenient. The solid ingredients are introduced; the cylinder is closed, evacuated to 1 mm. or less, completely filled with the desired liquid by suction, and sealed By translational movement of the piston and the use of steam at

atmospheric pressure in the jacket, rapid dispersion without foaming can be effected.

2.4.2 Sols

Viscosity - The viscosity of agar and agaroid dispersions is markedly influenced by the type of raw material and the processing conditions employed. The relative viscosities of agar and Gracilaria agaroid at 1% and 1.5% concentration have been reported the viscosity of an agar dispersion at 45° is relatively constant from pH 4.5 to pH 9 and is not greatly affected by age or ionic strength within the limit of pH 6.0-8.0. Once gelation begins, however, viscosity at constant temperature increases with time.

Gelation temperature - Agar is unique among polysaccharides in that gelation occurs at a temperature relatively for below the gel-melting temperature. Many uses of agar depend upon this high hysteresis. Agars and agaroids from different species have markedly different gelation temperatures, each of which is practically constant, with the exception of the agaroid from American Gracilaria confervoides, with exhibits seasonal variations of more than 20°. A gum from Hypnea musciformis, in which gelation temperature can be increased 60° by the addition of potassium chloride, is known.

The gelation temperature of agarose (agaran)sols is correlated with the methoxyl content of the gum (16).

Coagulants - Agar is quantitatively flocculated in the presence of electrolytes by ten volumes of ethanol, 2-propanol, or acetone. It is salted out by near saturation with sodium sulfate, magnesium sulfate, or ammonium sulfate. Prior to drying, such flucculated agar exists in metastable state, in which it is dispersible in cold water and in other solvents. In general, the higher the temperature of flocculation and the higher the concentration of electrolytes, the less soluble is the floc.

Many quaternary ammonium compounds cause turbidity and agaro pectin precipitation, as does silicotungstic acid. The most sensitive precipitants for agar appear to be tannic, phosphotungstic, and phosphomolybdic acids when used at pH 1.5-2.5

Miscellaneous - Viscosity, diffraction, and gel strength studies show that the chain length of agar is reduced by ultrasonic vibrations and strong gamma radiation, as well as by intensive agitation and high temperature. The infrared absorption spectra of agar, agarose (agaran), and other gums have been published.

2.4.3 Gels

Agar and agarose (agaran) are among the most patent gel-forming agents known, for gelation is perceptible at concentration as low as 0.04%. Threshold gels are valuable for their protective action, diffusion prevention, and texture

enhancement effects. Stronger gels are of valuable because of their strength, resilience, elasticity, relative transparency, relative permanence, and reversibility. Agarose gels are firmer but less elastic than gels from the parent agar.

Melting temperature - The melting temperature of an agar gel is a function of concentration and molecular weight Agar and agaroid gels with 1.5% solids melt from 60° to 97°.

Strength - The threshold gel concentration (TGC) is determined by the concentration of agar solids necessary for the formation of a particular gel under standard conditions The results correlated well with emulsifying, stabilizing,and protective ability and with gel strength in the 0.2-2.0% range.

Rupture at constant stress (cs) Method: In Japan, the Nikkankyo method has been used in afficial grading. Ten plungers loaded in an ascending series of mass are simultaneously lowered on ten replicate samples of a 1.5% gel (air-dry basis) that have been aged 15 hr at 20°. The maximum stress in g/cm^2 with stood for 20 sec without rupture is reported as the solidity of the sample.

Stress-Strain curve (ssc) method: This development from Stoloff's method yields information on gel strength, tenacity, and resilience. Screw-topped, 65-ml, ointment jars are filled to the 50 ml level with a 1.6% solids sol at 45°, sealed and held 1 hr in a stirred water bath maintained at

19.5°- 20.5°. A 1-cm^2, cylindrical plunger is pressed into the gel without lateral movement at 2 mm/sec. Load and depth of gel depression are simultaneously recorded to rupture. Rupture stress is a measure of strength; depression depth at failure varies as tenacity, and curves slope is a function of elasticity.

In the United states, three gel samples with 1.5% of solids aged 15 hr at 4° are subjected to different stresses estimated from previous ssc determinations and the rupture times are graphed. The stress in g/cm^2 required to effect rupture in 20 sec is taken from the curve.

Compatibility - Near neutrality, agar is compatible with most other polysaccharide gums and with proteins in the sense that flocculation or marked degradation does not occur when their dispersions are mixed. An exception is gum kino.

Near pH 3, flocculation occurs when warm agar and gelatin dispersions are mixed. Such mixtures are used to excellent advantage, however, in sherbets and ices by delaying the additions of acids until freezing has begun.

Sodium alginate and starch decrease the strength of agar gels, whereas dextrin and sucrose cause increases. Locust bean gum has a marked synergistic effect on the strength of agar gels. The incorporation of 0.15% of locust bean gum can increase the rupture strain of an agar gel 50-200%, depending on conditions. Iceland mass extractive (lichenan) and

carboxyethyl cellulose show similar action to a lower degree. Gelatin, Russian isinglass, and gum karaya tend to weaken agar gels, but only slightly. When added to warm agar dispersions, most salts, glycerol, sorbitol, the alkanalamines, and 1,2,6-hexanetriol have little effect on the strength of the gels.

Miscellaneous - Pure agar-water gels are rather stable. Those made with high-strength agar appear to be as stable as dry agar it self sterile and hermetically stored. Low-strength agars, however, deteriorate more rapidly in the gel than in the solid form. The fact that few microorganisms metabolize agar or elaborate enzymes that degrade it might explain the generally greater stability of agar gels in comparison with gels of other natural colloids.

Agar gels age slightly. After 1 hr at 25°, the strength increases about 1%/hr for about 8 hr, then increase at a decreasing rate and becomes stable in 15hr. Agar gels have micelle structure, and their elasliticity is energy elasticity rather than entropy clasticity.

2.5 Present Applications

2.5.1 Microbiology

Agar is most valuable in microbiology. The ideal agar is low in metabolizable or inhibitory substances, debris, and

thermoduric spores; has a gelation temperature of 35°- 40° and a gel-melting temperature of 75°- 85°; is readily soluble, and has good gel firmness, resilience, clarity and stability. Agar concentrations of 1-2% are commonly used for this purpose.

The Society of American Bacteriologists (now the American Society for Microbiology) have adapted specifications for agar for microbiological use. The latest (1958) requirements (17) are shown in Table 2-2.

2.5.2 Foods

Although agar is practically indigestible (18), it is used in many food products where its emulsifying, stabilizing, and gelling properties and the heat resistance of its gels are useful.

The uses of agar in the Food Industry are shown in Table 2-3 (13,19).

2.5.3 Medicine and pharmaceuticals

Agar has been widely used as a laxative for several decades. Medicinal-type agar especially prepared in the form of thin flakes designed to prevent the formation of obstructive masses and absorb 12-15 times its weight of fluid is well received professionally.

Agar is used as a suspending agent for barium sulfate in radiology, as an ingredient of slow-release capsules, in suppositories, in surgical lubricants, in emulsions of many types, and as a carrier of topical medicaments. It is used as

Table 2-2 The latest requirements for agar for microbiological use.

	Maximum	Minimum
Total solids	-	78%
Solubility, cold	2.0%	-
Solubility, hot	-	99.8%
Gelation temperature, 1.5%	39°	33°
Gelmelting temperature, 1.5%	-	70°
Rate of dissolution, 1.5%	15 min	-
Sol turbidity, 1.5%	10 ppm	-
Threshold gel concentration	0.25%	-
Protein nitrogen	0.32%	-
Reducing substances as galactose	10%	-
Chlorides as sodium chloride	1.5%	-
Viable spores	3/g	-
Debris count	30/g	-

Table 2-3 Representative Uses of Agar in the Food Industry.

Dairy Products	Bakery Product	Meat,Fish & Poultry	Miscellaneou
Ice cream stabilizer	Bread doughs	Sausage casing	Health foods
Ice milk	Cake batters	Pickled meats	Preserves
Milk shake	Pie fillings	canned tuna	Soups
Sherbets	Bakery jellies	antibiotic ice	Dessert water
Ice popsand water ices	Doughnut glaze	Preservative-	-gels
Cooked puddings	Flat icings	-meat coat	
Cream cheese	Meringues		
Yogurt	Cookies		
Packageable milk/ cream	Cake fillings		

a disintegrating agent and an excipient in tablets.

2.5.4 Miscellaneous

Agar has been found suitable for use in photographic stripping films and papers when esterified with succinic or phthalic anhydride and after enzymic hydrolysis.

The agar: is also used in solidified alcohol fuel, dyed coatings of paper, textiles and metals, laboratory. Agar is an ingredient of some cosmetic creams and lotions. As a corrosion inhibitor for aluminium.

Some newer uses for agar and its derivatives include agar as a setting inhibitor for deep-well cements; agar as an adhesive in gloss finishing of paper products (20), agar as an in-gredient in Fe - Ni - Cr - Ti electrolytes in the alloy - plating of steel, agar as an iron corrosion inhibitor in citric and malic acid solutions, and agar as an inhibitor of corrosion of iron and lead by distilled water.

Table 2-4 Costs of Imported Agar-agar in Thailand

	Code 130330 (AGAR-AGAR)	
	quantity (kg)	CIF Value (baht)
Jan - Dec 1970	173,933	14,839,938
Jan - Dec 1971	153,811	9,438,638
Jan - Dec 1972	179,979	11,953,630
Jan - Dec 1973	66,292	6,258,322
Jan - Dec 1974	102,923	13,619,678
Jan - Dec 1975	108,723	23,486,627
Jan - Dec 1976	181,138	37,386,551
Jan - Nov 1977	198,017	41,551,336