1.1 THE OBJECT :

The object is to study ion exchange resin - alkaloid systems and their usefulness towards practical application.

1.2 SYNTHETIC ION EXCHANGE RESINS : (1-10)

Ion exchange materials are of a wide variety. These may be inorganic or organic and of different shape and size. The common general structural principle is a framework with electric surplus charge and mobile counter ions. However, various types of materials behave markedly differently. Organic ion exchange resins are the most significant of the ion exchange materials. These are gels and the matrix consists of an irregular, macromolecular, three dimensional network of hydrocarbon chains. The ionic groups attached to the matrix may be of various types such as,

$$-SO_3^- \qquad -COO^- \qquad -PO_3^{-2} \qquad AsO_3^{-2}$$

for cation exchangers and

$$-NH_3^+ \qquad >NH_2^+ \qquad \rangle N^+ \langle \qquad \rangle S^+$$

for anion exchangers. Hence, ion exchange resins are crosslinked polyelectrolytes. They are insoluble, but have a limited swelling in water, depending on the crosslinking. The ion exchange behaviour of the resins is mainly dependent on the nature of the fixed ionic groups.

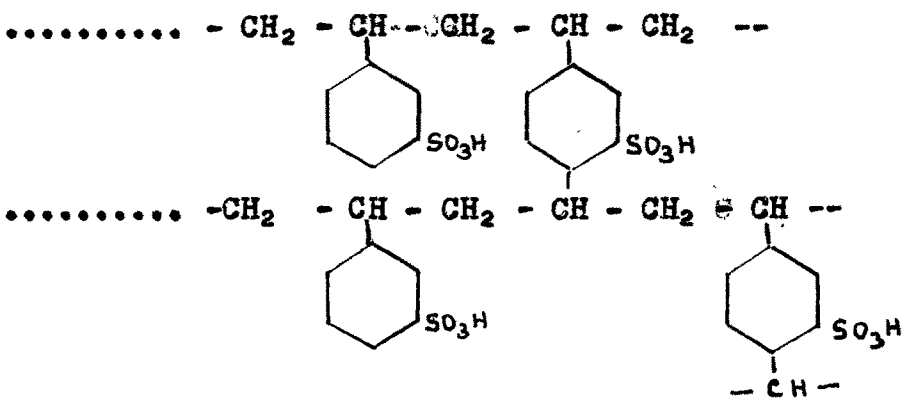
Commercial ion exchangers are insoluble solids. Recently liquid ion exchangers, such as long chain aliphatic amines and fatty acids or alkyl phosphates have become of great interest.

Organic synthetic resins in general, have superior chemical and mechanical stability, exchange capacity, exchange rates and versatility. Inorganic materials possess better thermal stability and resistance to radiation.

A wide variety of organic ion exchange resins have been prepared and some are available commercially under various trade names. The synthesis of ion exchange resin should yield a threedimensional, crosslinked matrix of hydrocarbon chains carrying ionic groups. The resins can be prepared by condensation polymerisation or addition polymerisation and the ionogenic groups can be introduced, before, during or after the polymerisation. These groups may be of one or more types giving monofunctional or multifunctional cation exchange or anion exchange or amphoteric resins.

Most of the earlier cation exchange resins were condensation products of phenol derivatives and aldehydes. Most of the present commercial resins are addition polymers prepared from vinyl monomers. These resins have a better chemical and thermal stability than the condensation polymers and their degree of crosslinking and particle size can be more easily controlled.

The monofunctional sulfonic acid cation exchange resins available are crosslinked polystyrenes with sulfonic acid groups, introduced by sulfonation of the polymer. The crosslinking agent used is divinylbenzene. Amberlite IR-120, Dowex 50, Nalcite HCR, Permutit Q, Duolite C-20 and C-25 and Lewalit S-100 are resins of this type. The structure may be imagined as :



Pure divinylbenzene is not readily available ; hence the resins are prepared with a commercial product consisting of a mixture of the different divinylbenzene isomers (about 40 to 55 %) and ethylstyrene (about 60 to 45 %). Ethylstyrene is also incorporated into the matrix. By varying the divinylbenzene content, the degree of crosslinking can be adjusted in a simple and reproducible manner. The nominal DVB content is used to indicate the degree of crosslinking ; it refers to mole percent of pure divinylbenzene (not of the commercial product) in the polymerisation mixture. Resins with low

degree of crosslinking, swell strongly and are soft and gelatinous. Resins with high DVB content swell much less and are tough and mechanically more stable. The copolymer beads are prepared by the pearl polymerisation technique. The monomers, from which stabilisers have been removed, are mixed and a polymerisation catalyst, such as benzoylperoxide is added. The mixture is then added to a thoroughly agitated aqueous solution at a required temperature (usually 85° to 100° C). The mixture forms small droplets, which remain suspended. A suspension stabiliser (gelatin, polyvinylalcohol etc.) in the aqueous phase prevents agglomeration of the droplets. The size of the droplets depends mainly on the nature of the stabiliser, the viscosity of the solution and the agitation and can be varied within wide limits. The polymer is obtained in the form of fairly uniform beads.

The sulfonation of the beads is simple, if proper precautions are taken. The cracking of beads may be avoided by first swelling the beads in an organic solvent such as toluene, nitrobenzene etc. It is advisable to transfer the sulfonated beads first to a highly concentrated electrolyte solution, which causes less swelling and then to dilute the solution stepwise. Sulfonation with concentrated sulfuric acid or chlorosulfonic acid results in practically complete monosulfonation of all the benzene rings.

Highly porous, macromolecular ion exchange

resins are prepared by a variation in the conventional pearl polymerisation technique. An organic solvent, which is a good solvent for the monomers, but a poor solvent for the polymer is added to the polymerisation mixture. As polymerisation progresses, the solvent is squeezed out by the growing copolymer regions. In this way, spherical beads are obtained with wide pores which permit access to the interior of the beads even when nonpolar solvents are used. The recent Amberlyst ion exchange resins are of this type.

Cation exchangers with specific preference for certain cations can be made by introducing groups which form strong complexes, preferably chelates with these cations. Resins with chelating iminodiacetic acid groups are now commercially available.

Most of the earlier anion exchange resins were condensation products of aromatic or aliphatic amines and aldehydes, dihaloparaffins or haloepoxides. Most of these contain weakly basic groups.

The more important anion exchangers are crosslinked polystyrenes, into which strongly or weakly basic groups are introduced by chloromethylation and subsequent amination. Reaction with tertiary alkylamines gives strong base quaternary ammonium groups and reaction with primary or secondary alkylamines or ammonia gives weak base amino groups. Anion exchangers with strong base quaternary phosphonium and tertiary sulfonium groups have also been prepared.

Amphoteric ion exchangers contain both acidic and basic groups. Snakecage polyelectrolytes are a novel variety of amphoteric resins. These are prepared from conventional ion exchangers by polymerisation of monomeric counter ions within the resin.

For specific purposes ion exchangers in the form of pellets, rods, belts etc. have been prepared by cementing ion exchange particles together with an inert binder or by impregnating suitable supporting carriers.

Ion exchange membranes have been prepared by various methods. The membranes may be homogeneous or heterogenous and have become of significant interest recently, particularly for desalting of water.

1.3 CINCHONA ALKALOIDS : (11)

Introduction :

Quinine is the most important of the cinchona alkaloids. In addition about 20 other alkaloids have been isolated from cinchona of which cinchonidine, quinidine and cinchonine are important. The alkaloids chiefly exist as salts of quinic and cinchotannic acids and their relative concentrations vary in different species. The bark which is known to the trade as druggist's bark has a quinine content of 1.8 to 2.0 %.

In the early years of planting, the total alkaloids were used for medicinal purposes under the

name of quinetum. In India quinetum was gradually replaced by cinchona febrifuge consisting of the residual alkaloids left after the removal of quinine. The Malaria Commission of the League of Nations redefined quinetum as a mixture of equal parts of quinine, cinchonidine and cinchonine and introduced a new product called totaquine or totaquina which is defined in the B.P. as containing not less than 70 % of crystallisable cinchona alkaloids-quinine, cinchonidine, cinchonine and quinidine of which not less than one fifth is quinine. Cinchona febrifuge varies greatly in physical character and composition, for use as an antimalarial drug. It should be of the same standard as totaquine.

Extraction :

The greater part of the world's production of cinchona barks is employed in the manufacture of quinine. For this purpose finely powdered bark is mixed with about one third of its weight of sifted slaked lime and a 5 % aqueous solution of caustic soda. The mixture is ex extracted under stirring in steam jacketed vessels, with high boiling kerosene. Three successive extractions are made. The mixed extracts are shaken with sufficient hot, dilute sulfuric acid to convert the alkaloids into sulfates. The oil is separated while hot and then neutral aqueous solution cooled when quinine sulfate separates out and is subsequently purified by recrystallisation

from aqueous solutions after decolorising with animal charcoal. The mother liquor containing the other alkaloids is treated with caustic soda and the precipitate of quinidine, cinchonidine and cinchonine extracted with dilute alcohol which dissolves the first two, leaving cinchonine behind ; the former two can then be separated by means of their neutral tartarates, that of quinidine being sonsiderably more soluble.

The method adopted by the Bureau of Science, Philippines, is to percolate to exhaustion with alcohol, a mixture of finely powdered bark, lime and water. The percolate is distilled to recover the alcohol, and the gummy residue treated with sulfuric acid to dissolve the alkaloids. The solution is decolorised with charcoal, filtered and the mixed alkaloids precipitated by the addition of sodium hydroxide.

An ion exchange process for the separation of alkaloids from cinchona barks poor in alkaloids has been developed and was successfully employed in the U.S.A. during the war period. This process was suggested for the recovery of alkaloids from the waste material left after the separation of the barks in India. The analysis of the alkaloids is based on processes such as polarimetry, colorimetry, turbidimetry, fluorometry and chromatography.

Uses :

The oldest and the most important use of quinine

is for the treatment of malarial fevers. Quinine continues to be effective inspite of its prolonged use. Quinine possesses marked bactericidal action and until the advent of sulfanilamide derivatives, quinine and certain of its derivatives were being employed in the treatment of bacterial infections. Quinine has been used as a sclerosing agent in the treatment of internal haemorrhoids and varicose veins. Quinine added to aquaphor, protects the skin against sun burn. Quinine sulfate is the most important salt of quinine used in therapy. Quinine ethyl carbonate and tannate are almost tasteless and are specially useful for children. Quinine with urea hydrochloride is used as a local anaesthetic. Practically tasteless compounds are obtained by combining quinine with an acid mixture derived from camphoric acid and an aromatic alcohol or a terpene alcohol or a phenol. In addition to their use in pharmacy, quinine and quinidine and their derivatives are utilised in insecticide compositions for the preservation of fur, feathers, wool felts and textiles. They are also ingredients of moth repelling preparations. Quinine sterate is used in hair lotions and pomades. The residual bark of quinine factories after the extraction of the alkaloids is a tanning material. Debarked cinchona poles are durable and resistant to termites. New and effective antimalarial drugs, specially, paludrine have certain advantages over quinine in the treatment of malaria. These new developments have no

doubt affected cinchona expansion schemes in India. However, from a strategic point of view, cinchona alkaloids are still of importance as indigenous materials particularly in war time, when imports may not be feasible.

1.4 ION EXCHANGE STUDIES WITH CINCHONA ALKALOIDS :

Ungerer (12) first examined the uptake of salts of quinine, cinchonine and strychnine on the calcium form of synthetic zeolites. Fink (13) took a patent for isolating cinchonine, strychnine and adrenaline from their aqueous extracts by a filtering material having adsorptive properties, such as asbestos and kaolin, cotton and asbestos or asbestos and kieselguhr. The pH of the solution was suitably adjusted to facilitate the separation. Applezweig (14) studied the removal of cinchona alkaloids by a cation exchanger of sulfonic acid type. Three possibilities were explored : (a) recovery of alkaloids from the mother liquor of the acid extracts of the bark after the major portion had been removed by alkaline precipitation, (b) purification of the crude totaquine obtained from alkaline precipitation and (c) application of ion exchange directly to the acid extracts of the bark in a cyclic system. Capacity determinations were carried out on a 200 cc. Zeo-karb column using quinine concentrations of 0.033 and 0.0033 M and flow rates of

approximately 5 and 50 cc./min. The capacity of a 200 cc. bed of Zeo-karb for quinine, from acid solution (1 % H_2SO_4) was found to be between 7 and 8 grams, before break through (Mayer's Reagent). To liberate the alkaloids from the column ammonical alcohol was used. Purification of totaquine prepared by alkaline precipitation of acid extracts of the bark was attempted by ion exchange. From 20 grams of totaquine precipitate, 2.5 grams of white crystalline material was obtained. Recoveries of totaquine from cinchona bark and scopolamine from datura plants were also effected by Sussman and others (15). The extract containing the alkaloids was brought into contact with a cation exchanger and then the cation exchanger was treated with aqueous alkali and a solvent. In a subsequent paper, Applezweig and Ronzone (16) described a portable unit for extracting usable antimalerial from freshly stripped cinchona bark in the field. Commercially dried cinchona bark was macerated with 0.1 N H_2SO_4. The acid was repeatedly cycled through a sulfonated coal cation exchanger and back into the maceration tank. The exchanger was regenerated with 0.5 N NaOH and stripped with alcohol, The crude alkaloid being recovered by evaporation. Rectified totaquine was obtained by precipitation from aqueous solution. An overall yield of 81.2 % within 82 hours was obtained. Mukherjee and Gupta (17) investigated the extraction of alkaloids from cinchona bark with hydrochloric acid and sulfuric acid over a range of acid

Concentrations and temperatures in the presence and absence of sodium chloride with Amberlite IR-100 and Ionac C-284. For elution, alkali was used and after an interval, alcohol was percolated. Ionac C-284 proved to be the best sorbent and showed highest elution effeciency but tended to soften and form a jelly in contact with alkali. Hence Zeo-karb, the next best and free from this defect was used. Applezweig (18) took a patent for the removal of quinine from a dilute solution in acid with Zeo-karb cation exchanger. The juice of the fresh material was passed through a column of Zeo-karb or Amberlite IR-100 or Ionac C-284. The sorbed alkaloids were eluted with ammonical ethanol. This method was used for the extraction of atropine, scopolamine and quinine alkaloids. Mukherjee and others (19) examined three cation exchange resins (Zeo-karb, Amberlite IR-100 and Ionac C-284) and two anion exchange resins (Deacidite and Ionac A-293) for the sorption of quinine sulfate, strychnine hydrochloride and other organic bases. The results showed that a resin having high sorption power for one alkaloid may not behave similarly with another alkaloid. The relative sorptive powers of the different resins, for each of the alkaloids studied were given.

Jindra (20) used an anion exchange resin of weakly basic type for the determination of several alkaloids. 0.1 to 0.2 grams of alkaloid salt was dissolved in 20 cc. of alcohol and passed through a

prepared column of Amberlite IR-4B. The flask and the column were washed with 50 cc. of alcohol at 50°C and the alkaloidal solution in alcohol was titrated with 0.1 N hydrochloric acid using a mixture of 10 drops of methyl red and two drops of methylene blue as indicator. The method was applied to quinine and cinchonine hydrochlorides and to a number of other alkaloids. Jindra and Pohorsky (21) have given detailed descriptions of the apparatus, reagents, preparations of the ion exchange columnsand general micro and semimicro methods of assay, applied to cinchona bark and other alkaloids. Bucke and Furrer (22) have described in detail, the determination of quinine and total alkaloids in the cinchona bark extracts by the use of ion exchange resins. In a subsequent paper they (23) found that sorption from cinchona bark was best with sulfuric acid extracts and the elution was best done with ethanol with or without addition of sodium hydroxide. A quantitative sorption occured within 14 hours by shaking the powdered cinchona bark and Duolite in dilute sulfuric acid but the subsequent separation of alkaloids from the resin was found to be difficult. Sanders and others (24) described an assay process using strongly basic anion exchange columns to separate quinine salts and ephedrine hydrochloride which was capable of giving results within 0.5 %. One and two column procedures are described. Yoshino and Sugihara (25) separated quinine and strychnine

chromatographically by sorption on weakly acidic cation exchange (NH_4-R) resins such as Duolite CS-101 or Amberlite IRC-50 and the subsequent elution with 0.1- 0.3 M ammonium chloride respectively. H-R exchanger could also be used but then a large amount of eluting solution was required to separate strychnine. Yoshino and others (26) classified some organic bases (a) quinine, cinchonine, ephedrine and berberine (b) nicotine and yohimbine (c) amino pyridine (d) antipyrine (e) acetanilide, caffeine, theobromine and theophylline, according to the facilities of being eluted with water from the cation exchange columns (sulfonic acid type and carboxylic acid type in the H and NH_4 form), on to which they had been sorbed. Street and Niyogi (27) separated a mixture of acetophenetidine, sulphacetamide, promacyine and quinine by a combination of chromatography and ionophoresis on cellulosic ion exchange sheets. Detection was accomplished by examination in ultraviolet light. Similarly (28) separation of a mixture of tablet fragments containing amobarbital, acetylsalicyclic acid, acetophenetidine, caffine, codeine and quinine into its constituent parts was accomplished by chromatography on modified cellulose ion exchange papers using both horizontal circular and ascending cylindrical paper chromatography. Street (29) has described a rapid method using ion exchange paper for the preliminary separation and detection of a mixture of quinine, strychnine and nicotine in whole blood.

Proteins were precipitated and the acid filtrate was extracted with ether. The aqueous phase was made alkaline with ammonium hydroxide and shaken with ether to extract basic compounds. The ether extract was evaporated to dryness and the residue was taken up in the chloroform. This solution was spotted on a cellulose cation exchange paper and subjected to chromatography in an aqueous solvent at pH 4.5. The separated compounds were detected by their fluorescence or absorbance in ultraviolet light at 254 mμ. Quinine showed a bright blue fluorescent spot and strychnine and nicotine as dark purple absorbing areas.

Saunders and Srivastava (30) studied the rates of sorption on, and elution from a carboxylic acid cation exchange resin for quinine. The factors which influenced the rate of sorption of quinine on Amberlite IRC-50 were found to be (a) the initial concentration of the solute (a millimoles /100 cc.) (b) the nature of solvent, sorption from 50 % ethanol solution being more rapid than that from pure ethanol for a given value of a (c) the method by which the resin was converted to the hydrogen form, aqueous 2 N acid producing a less effective absorbent than alcoholic 2 N acid (d) the amount of base already sorbed on the resin (x millimoles) (e) the stirring condition (f) the particle size of the resin and (g) the initial pH of the solution. An empirical relation $dx / dt = Ka (a - x) / x^2$

(where K = constant ; a = initial solution concentration in milimoles / 100 cc. ; x = milimoles sorbed by 5 grams of resin and t = time in hours), represented the rate of sorption quite closely for values upto 24 hours. The interaction of quinine with the resin was considered to be mainly a molecular sorption process. The sorption process has been visualised as a diffusion of base into the resin particle under chemical potential difference enhanced by the acid-base interaction with the resin and by the van der Waal's forces between the base molecule and gel structure of the resin ; the effect of viscous flow into spherical particles and the swelling of the resin caused the rate of sorption to fall off rapidly as the resin became saturated with base. Saunders and Srivastava (31) also examined the sorption of a number of organic bases from aqueous ethanol or ethanolic solutions by various carboxylic acid ion exchange resins and described the results of studies of the equilibrium distributions of some bases between the solutions and the resins. This, in the case of quinine has been demonstrated by showing that the distribution was independent of resin particle size and also that it was reversible. The systems studied have been classified into two groups. The first consisted of very weak bases which followed a simple distribution law, concentration of base in resin / concentration in equilibrium solution = constant. The second group, consisted of strong bases,

the distributions mostly followed a logarithmic law $Y = A \log C + B$ where C = concentration of base in the equilibrium solution in moles per liter of total phase volume and Y = corresponding base concentration in the resin phase in moles of base per liter of total resin phase volume. Observations have also been made of the swelling of the resins caused by saturating them with the different bases and it was found that swelling was a function of base sorbed.

Segal, Miller and Morton (32) have described the quinine form of a weak cation exchanger as an indicator for the determination of the presence of free hydrochloric acid in gastric juice without intubation. If a special cation (quinine) is combined with a cation exchange resin (Amberlite IRC-50 or XE-96) and the cation is displaceable only or mainly by hydrogen ion, is readily sorbed from the stomach and detectable in the urine or blood, the presence of free hydrochloric acid in gastric juice can be detected if quinine appears in the urine within 2 hours, after introduction of the complex, without subjecting the patient to intubation. Shay and other (33) found that the results of studies in patients after subtotal gastric resection indicated that the tubeless method for detection of the presence of free hydrochloric acid in the remaining gastric pouch was not suitable in these patients because of the rapid emptying of the quininium resin from the pouch. In such patients the

determination of pH of gastric contents during fractional gastric analysis was the best method for studying gastric acidity. Kamp (34) determined quinine together with caffeine and strychnine in cola sirup by ion exchange resin.

1.5 SUMMARY OF THE EARLIER WORK DONE IN THIS LABORATORY: (35,36)

1. The sulfates of four cinchona alkaloids (as $Q_2 \cdot H_2SO_4 \cdot nH_2O$, where-Q is the alkaloid base), quinine, quinidine, cinchonine and cinchonidine, were studied for their exchange behaviour with styrene divinylbenzene copolymer based sulfonic acid cation exchange resins. The variables studied were the relative degree of crosslinking of the resin, the particle size of the resin and the ratio of resin concentration to alkaloid sulfate concentration. The behaviour noted with alkaloid sulfate was of different type as compared to that of simple alkali cations (37). For resins of higher degree of crosslinking, the same fraction of the resin capacity is exchanged irrespective of the ratio of resin concentration to the alkaloid sulfate concentration This value varies with the relative degree of crosslinking of the resin. For low crosslinked resins, the effective exchange capacity is, to a small extent, dependent on the ratio of the resin concentration to

alkaloid sulfate concentration (38).

2. Several analytical methods for the estimation of alkaloid sulfates in dilute aqueous solution in the absence and presence of sulfuric acid were explored.

3. Ultraviolet absorption studies were made for aqueous solutions of cinchona alkaloid sulfates (quinine, quinidine, cinchonine and cinchonidine) of different pH (adjusted by addition of sulfuric acid or sodium hydroxide) and the ultraviolet absorption at invarient wavelengths was used for the estimation of the alkaloid sulfate in dilute aqueous solution (39).

4. Exchange equilibbia of quinine, quinidine, cinchonine and cinchonidine sulfates in the presence of added sulfuric acid of different concentrations were studied with the Amberlite IR-200, Amberlyst -15, Dowex 50-X4 and Dowex 50-X8 resins.

5. The uptake of the four bases from alcoholic and aqueous alcoholic solutions was also studied.

6. The exchange rates for dilute aqueous solution of quinine, quinidine, cinchonine and cinchonidine sulfates with various resins were studied at 35 and 45 ^{o}C. A simplified procedure was adopted by applying the second order law to the exchange process. For low crosslinked resins the rate of exchange, over a good part of the exchange reaction, was uniform and the rate constants were evaluated. For higher crosslinked resins, the exchange was by two exchange rates ; one relatively

fast and of shorter duration, and the other relatively slow and of longer duration. The second order law was applied to the slower rate and the relative rate constants were evaluated. The rate constants were a function of relative degree of crosslinking and of particle size of the resins. The temperature coefficients and the apparent energies of activation were evaluated for the four alkaloid sulfates.

7. A preliminary study of the column behaviour of quinine and cinchonine sulfates in aqueous solution with the different resins was carried out. Elution was studied with aqueous sulfuric acid of different concentrations and then by liberation of the base with caustic soda solution and elution with distilled ethanol.

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