Selectivity of Carbohydrate Films as Influenced by their Moisture Content

When droplets of a vegetable juice or extract dry sufficiently rapidly, a dry film may form around the droplet that is permeable to water but virtually impermeable to aroma components[1]. This has been confirmed in experiments in which an aqueous carbohydrate solution, to which 10 ppm acetone or another volatile substance as an aroma substance had been added, was used as a model juice[2]. Such a selective membrane may form not only around a droplet in a spray-drier, for instance, but also over a layer of a solution which is evaporating sufficiently rapidly[3].

This film is selective only if its moisture content is low enough. In fact it has been found that under certain conditions acetone cannot diffuse in the carbohydrate mixture used, if the moisture content of this mixture is less than 9%[2]. This threshold value of 9% moisture content was found by measuring the sorption of acetone and of ethyl acetate by the carbohydrate mixture at various moisture contents. Only powder with a moisture content above 9% sorbed acetone and ethyl acetate.

The phenomenon involved here is a particular case of 'liquid permeation' or 'pervaporation', a process for the separation of liquid mixtures by evaporation through a selective membrane. It is generally accepted that, in this process, the selectivity of the membrane is caused by differences in the solubilities and/or the diffusion coefficients[4-8]. In agreement with this theory, BROOKS[9] ascribes the retention of ethyl caprylate in aqueous gum arabic after spray-drying to the considerable difference between the solubility of water and of the ester in the almost dry film of gum arabic around the drying droplet.

Water happens to be one of the very few substances which very readily dissolve in hydrophilic carbohydrates and proteins. Therefore the aroma components will, in general, not permeate into films primarily composed of these hydrophilic compounds so easily as water does. Therefore it seems to us quite plausible to explain the selectivity of these dry films by a difference in solubility. But this in no way explains the above-mentioned threshold value of 9% moisture content. In order to learn more about this phenomenon, a number of sorption curves of volatile substances have been measured on the same mixture of carbohydrates using the method described above for the sorption of acetone and ethyl acetate[2].

The sorption curves were measured on malto-dextrin, type T, ex Messrs. Scholten, Foxhol, The Netherlands. According to the manufacturer its composition is: glucose 2.5%, maltose 27.0%, oligo saccharides larger than maltose 6.5%, non-reducing malto-dextrins 59.5%, minerals 0.5%, moisture 4.0%.

A 50% aqueous solution of this compound was first prepared. In order to get an easily dosable powder, this solution was mixed with chromosorb (W, non-acid washed, mesh size 60/100) in the ratio by weight of 1 part chromosorb to 1 part dry malto-dextrin. This mixture was then dried to the desired moisture content in a vacuum evaporator rotating in water at a temperature of 60–80 °C. An amount of 3 g of the powder so obtained was placed for 72 h in a stoppered bottle of ca. 50 ml, which also contained 10 μl of a volatile substance on filter paper. The amount of the volatile sorbed by the powder was then determined. The chromosorb was found to have no influence on the measurements with acetone; this was not checked in the case of the other volatile substances.

The volatile liquids tested were acetone, ethanol, 1-propanol, 1-butanol, methanol, ethyl acetate and carbon tetrachloride. The measuring of the amount sorbed by

[1] H. A. C. THIJSSEN, Inaugural Address, Technical University Eindhoven, The Netherlands (1965).
[2] L. C. MENTING and B. HOOGSTAD, Fd Res. *32* (1), 87 (1967).
[3] P. DE GRUYTER and N. V. ZOON, Werkwijze voor het verwijderen van water door een selektieve permeatie en verdamping (Process for removal of water by selective permeation and evaporation). Dutch patent application No. 6400502, 1965.
[4] A. S. MICHAELS, Lecture presented at the symposium on membrane processes for industry, sponsored by Southern Research Institute, Birmingham, Alabama, May 19–20, 1966.
[5] J. W. CARTER and B. JAGANNADHASWAMY, Br. chem. Engng. *9*, 523 (1964).
[6] B. H. SANDERS and C. Y. CHOO, Petrol. Refiner *39* (6) 133 (1960).
[7] N. N. LI, R. B. LONG and E. J. HENLY, Ind. Engng. Chem. ind. (int.) Edn *57* (9), 18 (1965).
[8] R. B. LONG, Ind. Engng. Chem. Fundamentals *4* (4), 445 (1965).
[9] R. BROOKS, Bgham Univ. Chem. Engr. *16*, 11 (1965).

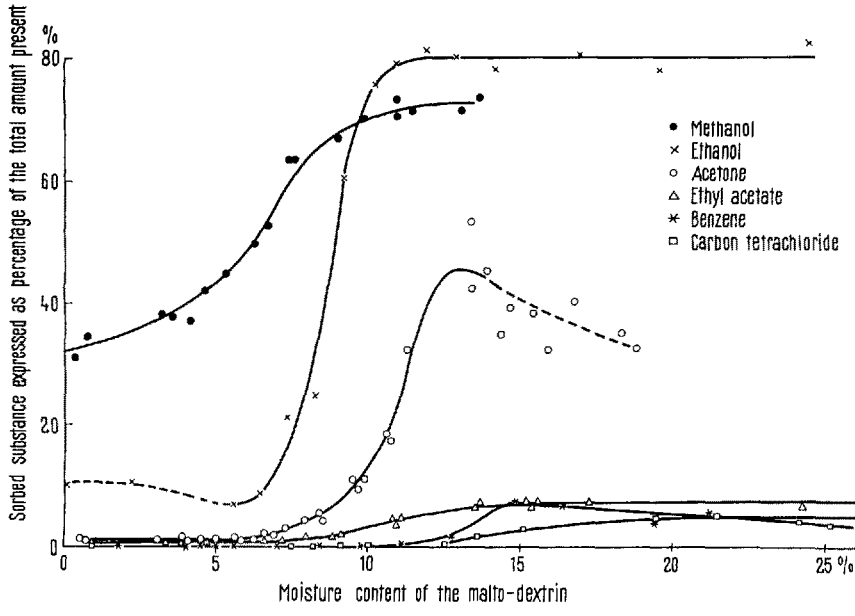

Fig. 1. Amount of volatile taken up by malto-dextrin mixed with chromosorb as a function of the moisture content.

the malto-dextrin in these experiments was greatly simplified by the use of compounds labelled with ^{14}C (ex Radiochemical Centre, Amersham, England). For this, 0.5 g of the samples of malto-dextrin were dissolved in 1 ml water after the sorption experiment. Subsequently 10 ml of water-miscible scintillator was added and the resulting solution counted with a Packard liquid scintillation counter, as already described in a previous publication[2]. Measurements were also made with camphor, the determination of which was carried out by gas chromatography.

The experiments were carried out at room temperature (ca. 20 °C). Moisture content is invariably expressed as a percentage of the total weight.

The amounts of the volatile substances sorbed by the malto-dextrin are represented in Figures 1 and 2 as functions of the moisture content. It will be seen that methanol and ethanol are still sorbed by the powder down to very low moisture contents. But this does not necessarily mean that methanol and ethanol are able to diffuse into the dry malto-dextrin; it may be that adsorption to or condensation on the surface of the powder occurs. As far as the sorption curve of ethanol is concerned, it is in any case certain that the dotted part of the curve in Figure 1 represents a range in which the sorbed ethanol is present only on the outer surface of the powder. This sorption curve was also measured on a continuous layer of malto-dextrin with a thickness of about 3 mm. After 1 week in a stoppered bottle which also contained 50 μl ethanol, the bottle was opened and a current of air was passed over the layer for 2 min to evaporate the ethanol on the outer surface. The ethanol content of the layer of malto-dextrin was then determined. The sorption curve (Figure 3) was now found to have the same shape as the curve for acetone. A sorption curve of methanol, measured in the same way had the same shape as the methanol curve in Figure 1.

Finally the sorption of 1-propanol and 1-butanol was measured. These 2 sorption curves resembled very much the curve for ethanol.

In Figures 1 and 2 we see that the moisture content at which the sorption curves begin to ascend steeply is dependent on the nature of the volatile substance. The moisture content at which this ascent begins cannot be determined accurately. Much more accurately determinable is the moisture content at which the sorption curve reaches its half-maximum height (half sum of minimum and maximum). We call this moisture content the 'moisture limit' (see Figure 2). For the sorption curve of ethanol, the minimum is taken from Figure 3. In Figure 4, this moisture limit is plotted as a function of the diameter of the molecule of the volatile compound. This diameter was measured on molecule models, built up of atom models (Courtauld's Maidenhead Laboratory and Griffin and George Ltd.). As 'diameter' we took the side of the smallest possible square opening through which the molecule model could just pass. From Figure 4 it will be seen that we found an almost linear relation between the above-defined molecule 'diameter' of the volatile substance and the moisture limit, irrespective of other molecule properties such as polarity and molecular weight.

We would rather not draw any further conclusions from the trend of the sorption curves at moisture contents above the moisture limit. During the determination of the amount of sorbed volatile substance in the malto-dextrin powder, this powder was in the open air for roughly 1 min, and in this time some of the sorbed substance may have evaporated. This error at moisture contents above the moisture limit cannot be neglected, especially when extremely volatile substances are used, for the diffusion rate in the malto-dextrin increases sharply with increasing moisture content.

The marked ascent in the sorption curves at a certain moisture content, as seen in Figures 1 and 2, and the increase of the moisture limit with the diameter of the molecules of the volatile compounds, as seen in Figure 4, lead us strongly to suspect that the malto-dextrin acts as a sieve, the mesh of which increases with increasing moisture content. We imagine the 'sieve' openings to be

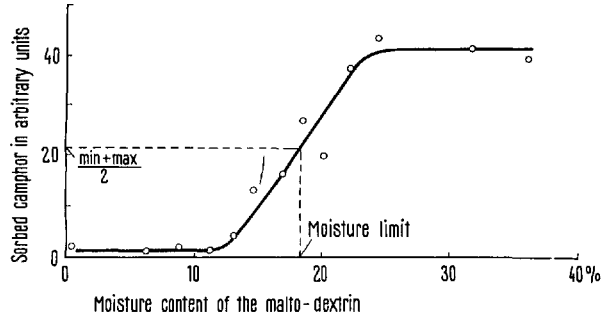

Fig. 2. Amount of camphor taken up by malto-dextrin mixed with chromosorb as a function of the moisture content.

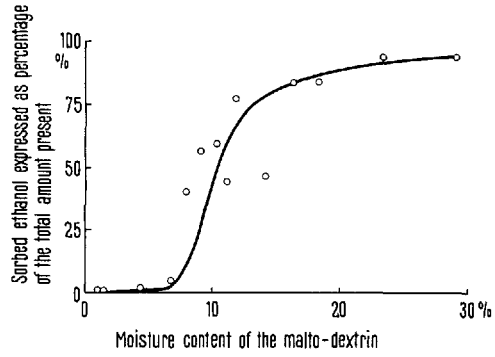

Fig. 3. Amount of ethanol taken up by a thick layer of malto-dextrin.

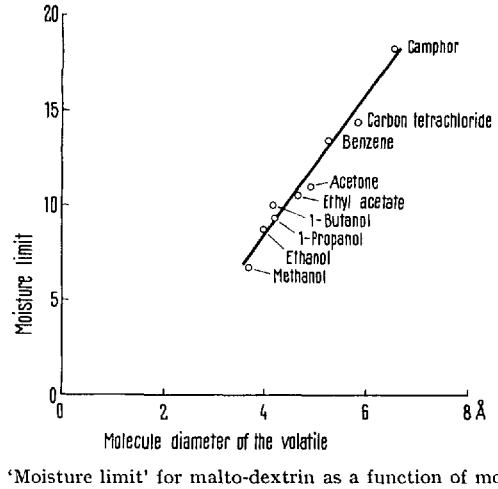

Fig. 4. 'Moisture limit' for malto-dextrin as a function of molecular 'diameter' of the volatile.

the interstitial spaces (moving openings or channels) between the large carbohydrate molecules, chiefly filled with the small water molecules. That these interstitial spaces very probably fluctuate as a result of diffusion of the water molecules and of the heat vibration of carbohydrate molecules which are partially bound together, does not in any way invalidate this sieve principle. At the same time this might explain why no clear coupling has been observed between water vapour transport and aroma transport through such selective polymeric membranes[10,11].

Zusammenfassung. Der Einfluss des Wassergehaltes auf die beim Trocknen pflanzlicher Säfte und Extrakte an der Flüssigkeitsoberfläche sich bildende, nahezu trockene, hydrophile Haut wurde untersucht.

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Unilever Research Laboratory Duiven, Zevenaar (The Netherlands), 28th March 1967.

[10] H. A. C. THIJSSEN and L. R. W. A. MIDDELBERG, Concentration and purification by freezing. International institute of refrigeration. Meeting of Commission 6-B; Delft, The Netherlands (1966).

[11] The authors are indebted to Prof. H. A. C. THIJSSEN of the Technical University Eindhoven for his helpful suggestions and to Mr. G. K. KOCH of Unilever Research Laboratory Vlaardingen for his help with the radioactive measurements.

Estimation of Free Glutamic Acid in the Differentiating Central Nervous System of the Chick

Glutamic acid is known to play an important role in morphogenesis[1] and metabolism[2] of the vertebrate central nervous system. For this reason, a quantitative survey of free glutamic acid concentrations in the 4 principal sectors (viz. fore-, mid-, hindbrain and the spinal cord) of the differentiating central nervous system of White Leghorn chick embryos at 4–20 days of incubation was made.

The estimation of the amino acid from the carefully weighed fresh tissue was made by the method of maximum colour density[3] resulting from the interactions of ninhydrin and the amino acid on a two-dimensional paper chromatogram (Whatman paper No. 1), with water-saturated phenol and the butanol-acetic acid-water (4:1:1) as the first and second solvent respectively at a constant temperature of 21 °C. The colour density was measured in a photovolt densitometer (Photovolt Corp., N.Y., USA; Model No. 501 A). The quantity of glutamic acid was found by comparing the readings of the measured quantity of the unknown samples with the standard curve prepared from a pure sample of glutamic acid by an identical method.

Observations and discussions. The concentrations of the glutamic acid in the tissue are shown in the Table. Though we had little success in tracing the appearance of adult-like amino acid patterns in many tissues, the embryonic brain of some amphibians[4] and of rats[5] shows high concentrations of glutamine and glutamic acid like those of the adult. Similarly, the varying concentrations of glutamic acid, as evidenced by the present study, may be correlated to a great extent with the epigenesis of the central nervous system in the chick. It may be observed from the Table that the first period of high glutamic acid concentrations occurs between the 4th and 8th day, and on the 10th day there is a decline in the quantity. In the chick, the differentiation of the central nervous system begins first in the spinal cord and last in the forebrain region[6], and the process is primarily complete mostly after 9 or 10 days' incubation[7]. During this time there is extensive cell proliferation, migration, degeneration and histogenesis. In this process, apart from playing a structural role, glutamic acid contributes much to (1) the supply of energy[8], (2) the mediation of the entrance of ammonia into the amino acid pool and also (3) into the transamination system[9]. It thus maintains an equilibrium between the concentrations of amino acids and proteins of the nervous system – the synthesis of which has much to do with morphogenesis[10]. Early differentiation of the spinal cord explains the higher concentrations of glutamic acid in this area than that in the 4-day-old embryos. In the case of mid- and also hindbrain regions of 12-day-old embryos, further differentiations[7] (viz. appearance of some finer strata in the optic lobes) start from the 11th and subsequent days, while in the case of hindbrains differentiations of 'folia' begin on the 12th day of incubation when the quantity of glutamic acid also becomes increased in these 2 sectors. The enhancement of glutamic acid concentrations in the 18- and 20-day-old embryos'

Amount of free glutamic acid (μg/100 mg of tissue) in the differentiating central nervous system of chick

Type of tissue	Age of embryos, in days							
	4[a]	6	8	10	12	15	18	20
Forebrain[a]	21.44	31.33	29.07	18.13	5.89	5.40	23.21	30.19
Midbrain		36.49	31.72	2.33	4.94	4.10	28.86	22.15
Hindbrain		30.40	51.94	8.13	16.56	3.29	21.96	6.43
Spinal cord	26.87	30.97	43.33	35.84	18.69	10.04	18.87	39.24

[a] Entire brain was considered.

[1] P. B. KUTSKY, J. exp. Zool. *124*, 263 (1953).
[2] D. RICHTER, *Biochemistry of Developing Nervous System* (Academic Press, New York 1955).
[3] R. J. BLOCK, *A Manual of Paper Chromatography and Paper Electrophoresis* (Academic Press, New York 1958).
[4] E. M. DEUCHAR, Biol. Rev. *37*, 378 (1962).
[5] W. A. HIMWICH, Int. Rev. Neurobiol. *4*, 117 (1962).
[6] V. HAMBURGER, *Biochemistry of Developing Nervous System* (Academic Press, New York 1955), p. 52.
[7] A. L. ROMANOFF, *The Avian Embryos* (McMillan Co., New York 1960).
[8] C. O. HEBB, Int. Rev. Neurobiol. *1*, 166 (1959).
[9] A. MEISTER, Physiol. Rev. *36*, 103 (1956).
[10] J. BRACHET, J. cell. comp. Physiol. *60*, suppl. 1, 1 (1962).