DRUG EXTRACTION. I. A STUDY OF VARIOUS MENSTRUA FROM THE STANDPOINT OF SWELLING EFFECTS, PENETRATION AND EXTRACTION.

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(*Continued from page 901, September Journal.*)

Swelling of Powdered Belladonna Root.—Studies of the swelling of powdered drugs are obviously of importance, since this is the form in which drugs are extracted. Scoville (2) measured the swelling of a ground drug, 5 Gm. of gentian in about a No. 30 powder being placed in a specially graduated 25-cc. cylinder and the volume measured before and after treatment with a liquid, allowing time for settling. The error caused by the buoyant effect of the liquid on the drug particles was recognized by Scoville.

In the present study this method was improved by centrifuging the mixture of drug and solvent, thus overcoming the buoyant effect of the solvent and obtaining a rather complete and sharp separation of liquid and solid. Graduated centrifuge tubes of 15 cc. capacity were used in a large size centrifuge (Size 1, Type SB, operating at 5 amperes, 110 volts, 60 cycles, made by the International Equipment Co.). The centrifuge was run close to maximum speed, which was kept constant throughout the experiments. Except where otherwise specified, 0.50 Gm. of powdered drug and 8.0 cc. of liquid were used in the experiments, these amounts being found to be most practical. The mixtures of powdered drug and liquid were centrifuged for ten minutes which was the time ascertained by trial as necessary for complete packing. In stating the results, the ten minutes of centrifuging is included in the maceration time. The tests were conducted in duplicate, the volume of the powdered drug being read off on the graduated centrifuge tubes.

TABLE XVI.—SWELLING OF BELLADONNA ROOT (IN NO. 40 POWDER) IN VARIOUS MENSTRUA.

Menstrua. Volume of: Glycerin.	Alcohol.	Water.	Dry. 0.	Time of Maceration in Minutes. 10.	20.	40.	60.	120.	360.	720.	1440.
0	0	1	100	161	161	152	161	161	166	171	152
0	1	7	100	152	161	161	161	152	152	161	142
0	1	3	100	142	138	138	142	142	138	142	133
0	1	1	100	123	133	123	128	123	114	114	114
0	7	3	100	119	119	109	114	114	109	114	109
0	5	1	100	104	114	109	114	104	104	109	104
0	9	1	100	114	114	114	114	114	104	104	104
0	1	0	100	114	114	114	104	100	104	104	100
1	0	3	100	138	142	142	142	142	142	142	152
1	0	1	100	138	142	142	142	142	142	142	152
4	0	1	100	123	128	138	142	142	142	142	152
1	3	0	100	104	109	109	109	109	109	109	119
1	1	0	100	104	104	104	104	109	114	114	119
4	1	0	100	100	100	114	114	114	119	133	133
65	250	685	100	119	119	119	119	119	123	128	128
100	500	400	100	104	104	100	104	104	104	104	114
75	675	250	100	104	104	100	100	100	109	104	104

Swelling of Belladonna Root in Various Menstrua.—Through a reputable dealer and miller of crude drugs a 125-pound shipment of belladonna root U. S. P.

was obtained as follows: 10 lbs. whole root, 40 lbs. of 40 mesh and 25 lbs. each of 20, 60 and 80 mesh, prepared according to the following specifications: "The above to be prepared by taking 125 pounds of belladonna root, selecting a representative sample of 10 pounds for the whole root and a representative 40-lb. sample to be milled to 40 mesh and three 25-lb. samples to be milled to 20, 60 and 80 mesh, respectively. Each 25-lb. (or 40-lb.) portion is to be milled separately so that each portion will be as nearly alike as possible except for the difference in the milling." It was thought that by use of this 125-pound supply of the drug, all experiments would be strictly comparable. A thorough study of the shipment was made by C. L. Huyck, and it was found that the drug conformed to the U. S. P. requirements.

For concise presentation, the results in the tables which follow represent the average of two determinations, expressed on a percentage basis, the volume of the dry drug being taken as 100.

With alcohol-water mixtures the results indicate that the swelling decreases with increasing alcoholic content. The No. 40 powder reached a maximum swelling in the first ten minutes in the alcohol-water mixtures; the later noticeable fall in percentage swelling may be due to a softening of the drug after prolonged contact with the liquid and subsequent firmer packing by the centrifugal force.

In glycerin-water mixtures, the final swelling was the same regardless of whether the liquids were present in equal amounts, or whether the glycerin or water was in excess; however, the mixture of glycerin 4 vol.—water 1 vol. required a longer time for reaching maximum swelling. In glycerin-alcohol mixtures there was less swelling than in glycerin-water mixtures. In glycerin-alcohol-water mixtures the swelling after 24 hours was greater with the mixtures having the greater proportion of water.

Effect of Fineness of Powder on Swelling.—Using the official menstruum specified by the U. S. P. X for Fluidextract of Belladonna Root, *i. e.*, a mixture of alcohol 5 vol.—water 1 vol., tests were conducted by the centrifuge method on belladonna root in No. 20, 40, 60 and 80 powder.

TABLE XVII.—SWELLING OF BELLADONNA ROOT OF DIFFERENT DEGREES OF FINENESS IN A MIXTURE OF ALCOHOL 5 VOL.—WATER 1 VOL.

Powder No.	Dry. 0.	Time of Maceration in Minutes. 10.	20.	40.	60.	120.	360.	720.	1440.
20	100	113	117	113	113	113	130	130	117
40	100	100	104	109	109	104	104	114	104
60	100	100	100	100	100	105	105	105	105
80	100	105	110	115	115	115	115	115	115

The volume occupied by a given weight of powdered drug would naturally be greater for the coarser powders, since large particles pack less efficiently than small particles in a given space. The results in Table XVII indicate that for the No. 20, 40 and 60 powders, the larger the powder, the greater the apparent swelling; this may be due to the fact that as the larger particles swell, the packing becomes still less efficient. An opposing tendency, however, arises in the fact that swelling takes place largely near the surface, so that small particles should show a greater percentage swelling on the basis of this factor taken by itself. In the No. 80 powder, the greater swelling as compared with the No. 40 and 60 powders may be due to an overbalancing of the factor of efficiency of packing by the factor of the increased coefficient of swelling in smaller particles.

Effect of Proportion of Liquid in Maceration on Swelling.—This point was considered from the standpoint of the effect on percentage swelling in maceration preliminary to packing in a percolator. Tests have been carried out by the centrifuge method, by the use of an oedometer (3), and by a packing method using a graduate.

For tests of swelling of drug powders with small amounts of moistening liquid, a further study of methods is required, since the results obtained by the different techniques are contradictory to a certain extent; however, the results seem to indicate that maximum swelling is not attained with amounts of liquid ordinarily used in moistening drug powders preparatory to packing in a percolator.

Discussion of Results on the Swelling Effect of Solvents.—The drying of a drug represents primarily a removal of water, although other secondary changes occur. Tissues are originally more or less saturated with water, and a marked decrease in size results from drying, due perhaps in greatest measure to loss of turgidity and collapse from loss of water. From these considerations it would be natural to consider swelling as primarily a reversal of the drying process, with some differences due to the irreparable damage to the plant protoplasm during the drying. The hydration of such dried material (4) would take place as in other dead material by means of imbibition and adsorption, with osmosis by the action of vacuolar materials almost eliminated. This illustrates the botanical maxim that when a living cell dies, it loses its osmotic powers.

While water seems to be the natural swelling agent, some other liquids, such as alcohol, resemble water sufficiently to be taken up by the tissues. It would seem that, in order to cause swelling, a liquid must permeate the tissues and wet the inner surfaces. The swelling by absolute alcohol would be an example of this; it is possible that the momentary shrinkage caused by absolute alcohol is due to the abstraction of water from the tissues and that the swelling which follows is due to the gradual permeation of the cells by absolute alcohol. In the tests on swelling of strips of chestnut wood in absolute alcohol it was recognized that there was a possibility that the absolute alcohol on the slide might take up sufficient moisture from the air to affect the results. However, the tests on blocks of chestnut wood were carried out in stoppered and sealed flasks thus eliminating any possible effect of moisture from the air and it was thus established that the swelling effects are due to absolute alcohol itself.

The slower approach to equilibrium in the glycerin-water and alcohol-water mixtures might be thought of as due to a slower permeability of the cells to the alcohol-water and glycerin-water mixtures.

The fact that concentrated glycerin solutions cause only one per cent of swelling may be due to the stability of glycerin hydrates, or to a lack of permeability of the cells to concentrated glycerin solutions. The well-known affinity of glycerin for water is probably due in part to the formation of hydrates. Such water of hydration would not be available for hydration of the dried plant tissues unless these had a greater attraction for the water than the glycerin had, in other words the distribution of water between the tissues and the glycerin would depend upon the composition and relative stability of the respective hydrates. Glycerin caused a shrinkage of the blocks of chestnut wood, followed by recovery and gradual swelling. The results are reminiscent of the observation by Klebs (5) in 1888, that plant cells placed in a solution of glycerin first underwent plasmolysis and then gradually recovered, thus indicating gradual permeation by the glycerin.

Ethylene glycol, diethylene glycol and carbitol cause about the same percentage swelling as does water, except that water comes to equilibrium much more rapidly. Propylene glycol and dioxan caused less swelling than alcohol.

In general the results with the glycerin-water, glycerin-alcohol and alcohol-water mixtures indicate that the swelling in mixtures of two liquids is practically an average of the effect of the liquids themselves when allowance is made for the relative proportions of the two liquids in the mixture. An examination of the graphs (see Graphs 1 and 2) indicates that the general form of the swelling curves is very similar for alcohol-water and glycerin-water mixtures. However, there are some noteworthy differences, in that a smaller proportion of glycerin than of alcohol is required to bring about a decrease in the primary swelling and a lengthening of the period necessary for the attainment of equilibrium, and in that the addition of one part of water to 9 parts of glycerin has no effect on the swelling curve, but in the corresponding alcohol-water mixture there is a difference.

The results of the work on strips of chestnut wood indicated that a variation in p_H in aqueous solutions had no effect on the swelling, but in the work on blocks of chestnut wood it was found that $0.01N$ aqueous HCl and NaOH both slightly increased the swelling. In alcohol, swelling of woody tissues is decreased by acids, noticeably in $0.1N$ concentration. Alcoholic NaOH decreased the swelling of blocks of chestnut wood, while in the work with strips of chestnut wood alcoholic NaOH increased the swelling. The swelling with strips may be due to a weakening action of the NaOH on the thin sections used. MacDougal (4) also found that tissues swelled more in alkaline solutions; he states further that "acids decrease the hydration capacity of agar and of the pentosans which enter into the composition of certain types of plant cells." The effects of acid and alkali in alcoholic solution are noticeable particularly in cases where the alkali or acid are present in the alcohol when it is first added to the dry wood. If the wood has already been acted upon by the alcohol before the addition of alcoholic acid or alkali, the effect is less noticeable. After a tissue has been treated with alcoholic acid or alkali, treatment with alcohol is able to reverse the effect of the acid or alkali only partially. This result is in accord with the statement of MacDougal (4) that "no swelling agent yet tested has been found to reverse the action of another solution so fully as to bring the dimensions of the sections down to the dimensions which might have been attained in the second agent alone." However, the results obtained on successive additions of solvents show that water, alcohol and absolute alcohol each have a characteristic effect on the tissues and that this individual effect tends to assert itself regardless of whether the solvent is applied to a dried tissue or to one that has already been treated with another solvent.

Comparative Swelling with Grain and across Grain.—Current theories regarding the structure of the cell wall offer an explanation for the fact that swelling of wood is greater across the grain than with the grain. In 1858, Nägeli (6) proposed a theory that the cell wall consists of ultramicroscopic, crystalline, molecular complexes, which he called micellæ, and which were supposed to be arranged spirally. Likewise, in 1924 Koehler (7) stated that theoretically the cell walls are made up of ultramicroscopic fibrils running spirally in the cell walls, the water being held almost entirely between the fibrils. According to Schorger (8) each micella is normally surrounded by a film of water, but in the dry condition the micellæ are drawn together. On exposure to water, "swelling takes place up to the point where the pressure of the aqueous layers equals the cohesion between the micellæ."

On the basis of X-ray investigations, Sponsler (9) concluded that the structural units of the cell walls of fibres are $C_6H_{10}O_5$ molecules attached in chains running parallel to one another and very uniformly spaced. According to Sponsler and Dore (10) the $C_6H_{10}O_5$ molecules themselves are held together to form chains by bonds of primary valence, while the chains are associated and held together by bonds of secondary valence. They state that since the $C_6H_{10}O_5$ molecules are held together by primary valence, water molecules cannot penetrate between them and consequently longitudinal swelling of cellulose fibres cannot occur to any great extent. The swelling across grain is explained on the basis that water is able to penetrate between the chains, which are held together by secondary valence only. The water molecules become adsorbed upon the oxygen atoms, thus increasing the distance between the parallel chains and causing swelling.

Swelling of Fresh and Dried Woody Tissues.—There is practically no swelling of the fresh blocks of oak sapwood and of Elberta peach wood in liquids. However, the fresh oak sapwood absorbed 12 per cent of its weight of water and the fresh Elberta peach wood absorbed 42 per cent of its weight of water during the time of the experiment. This situation is explained on the basis that the liquids which enter open spaces in the wood do not cause swelling but merely occupy space which was previously filled with air. Thus Hawley and Wise (11) have stated that the water held in the microscopic cell cavities does not cause swelling of wood. According to Detmer (12) the mere filling of the lumina of the wood elements with water cannot bring about any increase in volume of the material, but only the liquid imbibed by the solid substance causes swelling. Detmer states that imbibition is by no means capillarity; a fluid entering by capillarity occupies previously existing spaces, while when imbibition takes place, the molecules of the liquid push in between the micelles actually making space for themselves and thus bringing about an increase in volume. These considerations make clear how a liquid may be absorbed in quantity in the open spaces in the plant tissues and yet may not cause appreciable swelling either because the micelles are already distended as in fresh wood or because the liquid does not have the properties facilitating imbibition as distinguished from capillarity. Steel (13) takes the view that when a substance absorbs a liquid without swelling, the cohesion of the molecules of the substance is too great to permit them to be forced apart by the liquid.

In studying the data concerning the effect of water and other liquids on blocks of woody tissues dried to constant weight, we find that recovery of original size occurs to a greater extent and at a more rapid rate with the blocks dried at room temperature than with the ones dried in an oven at 90° C. These results are in accord with the general rule of colloid chemistry that gels which have been partially dehydrated have a lowered capacity for reimbibing water. This has sometimes been explained on the basis that during drying the salts and other dissolved substances are finally left in a concentrated form on the tissue surfaces, and thus affect imbibition in a different manner than when diffused in dilute form.

Effect of Thickness of Woody Tissue on Swelling.—MacDougal (4) showed that in thin sections, the coefficient of expansion is relatively high and solvation is attained quickly. Our results are in agreement; thus the chestnut wood blocks increased only 7 per cent in thickness while the thin sections (0.05 mm.) increased 20 per cent in width. Cells at or near the surface of a section or block may swell until the forces causing swelling are counter-balanced by the elastic force of the stretched cell wall. Cells on the interior of a block, in order to swell, must overcome not only the elastic forces of their own cell walls but also the restrictions of the surrounding cells; in blocks possessing considerable rigidity it can be readily understood that the inner cells must swell less than the outer cells and thus the coefficient of expansion is relatively low in thick pieces or blocks. In a block the outer layers would cause the greater amount of swelling. This is in accord with our results, which show that the greater part of the swelling of blocks took place in considerably shorter time than was necessary for the liquids to penetrate to the center of the blocks. (*To be continued.*)

STUDIES ON STRYCHNINE. III. THE EFFECTIVENESS OF SUCROSE, SACCHARIN AND DULCIN IN MASKING THE BITTERNESS OF STRYCHNINE.[*,1]

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Earlier papers in this series (1, 2) on the masking of the bitter taste of strychnine gave the results of experiments with a number of chemically unrelated substances. The results obtained with 1.5 and 10 per cent sucrose solutions suggested the desirability of continuing and extending studies with sucrose as well as with other sweetening agents.

In the series of tests reported in this communication, strychnine alkaloid, sulphate and hydrochloride were obtained from several manufacturers. Chemical tests showed the purity of the products used. The sucrose was a sample of factory-run beet sugar. Soluble saccharin and *p*-phenetylurea (dulcin) were obtained from commercial sources. The technic used in our previous studies was followed here. Five cc. of a solution of strychnine and 5 cc. of a solution of the sweetening agent

* Scientific Section, Miami meeting, 1931.

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