The animals were then removed, the excess water wiped off, and the animals accurately weighed in a tared beaker to the first decimal place.

The solutions of strychnine sulphate and brucine sulphate for hypodermic injection were made up in sterile physiological saline solution, and the concentration so adjusted that each animal would receive approximately the same amount of fluid (about 0.5 cc.).

The fluid containing the alkaloidal salt was injected into the ventral lymph sac by means of a Luer type hypodermic syringe. A preliminary series of observations was conducted in order to arrive at the comparative convulsive dose of the drug. Observations were made continuously after the injection of the dose, and the time was noted when the animal became hypersensitive, and when the first convulsion occurred.

B.—RABBITS.

Rabbits used in the observations were obtained from local sources. The weight of each animal was ascertained in kilograms. The solutions of strychnine and brucine sulphates were made up in sterile, physiological saline solution, as described in the technique of the frog method. The dose was administered by subcutaneous injection in the abdominal region with a sterile hypodermic syringe, the concentration being adjusted so that each animal received approximately the same volume of fluid.

Observations were made constantly, and the time to hypersensitivity, and to the first convulsion were recorded.

C.—DOGS.

The weight of each dog was determined in kilograms, and the solutions of strychnine and brucine sulphate prepared in sterile, physiological saline solution, and injected by means of a sterile syringe subcutaneously in the abdominal region.

Observations were made constantly, and the time recorded when the animal appeared hypersensitive, and when the first convulsion occurred.

D.—CATS.

The same technique was employed with cats as has been described under "rabbits" and "dogs." Observations again were made constantly, and the length of time to hypersensitivity, and the time to the first convulsion were noted.

(To be continued)

A STUDY OF THE PREPARATION, QUALITATIVE DIFFERENTIATION TESTS, AND METHODS OF EVALUATION OF THE VARIETIES OF ALOE.*

A THESIS SUBMITTED TO THE FACULTY OF THE GRADUATE SCHOOL OF THE UNIVERSITY OF MINNESOTA.[1]

BY KARL GOLDNER.

Aloe, U. S. P. X (1), is the inspissated juice of the leaves of *Aloe Perryi* Baker, known in commerce as Socotrine Aloe; of *Aloe vera* Linné, known in commerce as

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[1] In partial fulfilment of the requirements for the degree of Master of Science.

Curacao Aloe; or of *Aloe ferox* Miller, known in commerce as Cape Aloe (*Fam. Liliaceæ*).

Aloe yields not more than 4 per cent of ash, not more than 10 per cent of moisture and not less than 50 per cent of water-soluble extractive.

Unground Socotrine Aloe occurs in yellowish brown to blackish brown, opaque, smooth and glistening masses; fractured surface somewhat conchoidal; odor characteristic.

Unground Curacao Aloe occurs in orange to blackish brown, opaque masses; fractured surface uneven, waxy, somewhat resinous; odor characteristic, disagreeable.

Unground Cape Aloe occurs in reddish brown masses, usually covered with a yellowish powder, or in thin, transparent fragments, of a reddish brown color; fracture smooth and glassy; odor characteristic, sour and disagreeable.

The taste of each variety of Aloe is nauseous and very bitter.

Powdered Aloe is yellowish brown to dark reddish brown; mounted in a bland, expressed oil, it appears as yellowish to reddish brown angular or irregular fragments, the depth of color depending to some extent on the thickness of the fragments.

TESTS FOR IDENTIFICATION AND DIFFERENTIATION.

Aloe is tested according to Bainbridge and Morrow (2), who observed that, upon addition of nitric acid, Socotrine Aloe gives a yellowish to reddish brown solution; Curacao Aloe a deep red solution; Cape Aloe a reddish brown solution, changing to purplish brown and finally to greenish.

About 1 Gm. of powdered aloe, accurately weighed, is mixed in a flask or bottle with 25 cc. of cold water, shaken occasionally during two hours, transferred to a filter, tared after having been dried over sulphuric acid, and washed with sufficient water to make the filtrate measure 100 cc. The residue on the filter, dried over sulphuric acid, should not exceed 50 per cent of the weight of powdered aloe taken. The color of the filtrate, viewed in the bulb of a 100-cc. volumetric flask, is light yellowish brown with Socotrine Aloe, reddish brown with Curacao Aloe, and yellowish with Cape Aloe. The filtrate darkens upon standing.

To 5 cc. of the filtrate mentioned above are added 2 cc. of nitric acid: the mixture is of a yellowish to yellowish brown color with Socotrine Aloe; a deep red color with Curacao Aloe; a reddish brown color, changing to vivid green, with Cape Aloe.

The filtrate mentioned above responds to Schonteten's test for the presence of aloin. To 5 cc. of the filtrate are added 45 cc. of water and 20 cc. of sodium borate solution (1 in 20): a greenish fluorescence results and, upon standing, the liquid acquires a brownish color.

The filtrate mentioned above responds to Bornträger's test. To 10 cc. of the filtrate are added 90 cc. of water and the solution shaken with 10 cc. of benzene: upon separating the benzene layer and adding to it 5 cc. of ammonia T. S., a permanent, deep rose color is produced in the lower layer. The color is dependent upon the presence of emodin.

Aloe should be free from gum or inorganic impurities. A mixture of 1 Gm. of Aloe and 50 cc. of alcohol are heated gently and then cooled: a nearly clear solution should be obtained.

Cripps and Dymond's tests (3) for the detection of aloe is made as follows:

One grain of the solid substance to be tested is treated in a porcelain dish, or in a glass mortar standing on white paper, with 16 drops strong sulphuric acid and triturated until the whole is dissolved. Four drops of nitric acid of 1.42 sp. gr. are then added, and this is followed by 1 oz. of distilled water, when, in the presence of aloe, a color will be produced varying from deep orange to crimson, according to the kind of aloe employed. The result is confirmed by adding ammonia, when the color is intensified, usually to a deep claret red. The test not only allows of the detection of aloe, but also gives a fair indication of the kind of aloe under examination.

Stacy (4) recommends the addition of a fresh solution of potassium ferrocyanide to a cold aqueous solution of aloe, when a pink color, which appears quickest in the boiling solution, is produced by 1 part in 10,000 of Barbadoes aloe. Socotrine or Cape aloe (1 part in 250) or commercial aloin, give green colors. No reaction is obtained in the presence of cascara sagrada or rhubarb. Previous extraction with petroleum spirit, benzene or chloroform, though not with ether or ethyl acetate, produces a negative reaction.

Klunge's test (5) consists in the addition, to a highly dilute and therefore nearly colorless solution of aloe, of 1 drop of copper sulphate solution, followed by the addition of sodium chloride and alcohol. On the addition of the copper sulphate, the original yellow color of the solution is intensified. The addition of the salt, however, followed by a little alcohol or warming, changes this to a red color, the intensity and permanence of which differs with the different varieties of aloe. The use of alcohol may be avoided if the solution is warmed, but the addition of a considerable amount, as recommended by Leger, has the advantage that it dissolves the flocculent precipitate caused by the salt.

The plant from which the drug aloe is obtained has stems about a meter high, which bear at the summit a cluster of thick, succulent leaves, lanceolate and spinous-toothed.

The methods of obtaining aloe commercially vary somewhat with the locality. The usual method (6) is to dig a hole in the ground and cover the bottom of it with a goat or horse hide. The leaves are cut with a curved knife and placed in a circle, the cut surfaces under and to the inside, to a height of one meter. By this method of packing, the cut surfaces of all layers can drain directly into the hole After several hours the leaves are removed and the sap poured into a container, often an empty petrol can. The juice is then thickened by boiling in iron tubs over a free fire by a rather carelessly conducted process. During the inspissation, the juice is stirred vigorously with a wide wooden spatula. The soft, warm mass is then poured into boxes, gourds (Barbadoes aloe), or sometimes monkey skins (Socotrine aloe.)

On the Island of Barbadoes the leaves are cut off at the flowering time and placed in "V" shaped troughs of which several empty into a container. The sap is allowed to flow for 1 to 2 hours, is collected in large reservoirs, and then evaporated in copper vessels. The foam is skimmed off as it forms.

CONSTITUENTS OF ALOE.

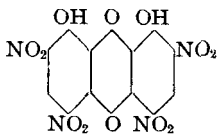
The most important constituent of aloe is aloin, which was first crystallized and named by the Edinburgh apothecaries Th. and H. Smith in 1851. Aloins from Barbadoes, Cape (7), Uganda (8), Zanzibar (9), Socotrine (9), Curacao (10), and Jafferabad (11) aloes are said to be identical. Aloin from Sicilian aloe and aloin from Natal aloe are said to be of different constitutions.

Barbaloin crystallizes with three molecules of water, which may be removed by drying at 110° in a current of hydrogen. When dry, it is very hygroscopic. It forms pale yellow needles, whose melting point varies according to the source of the aloin. The melting points of water-free aloins are given by Tschirch (12) as: Barb-, Cur-, Cape and Uganda aloin: 147°; Jafferabad: 152°; Socotrine: 157°; Zanzibar: 212°. α_D in aqueous solution: + 21.4°.

If barbaloin is heated with concentrated sulphuric acid and to an alcoholic solution of the residue is added an alcoholic solution of potassium cyanide, a violet color is formed which changes to a rose color (Formanek (13)). A drop of a solution of barbaloin in concentrated sulphuric acid, over which vapors of fuming nitric acid are blown, becomes blue in the center, violet on the edges. If the solution is

diluted with water, it becomes cherry-red; by the addition of sodium hydroxide solution, deep carmine red (Leger (14)). It is scarcely precipitated by lead acetate in excess, differentiating aloin from the oxymethyl-anthraquinones of rhubarb, frangula, etc. (Kremel (15)).

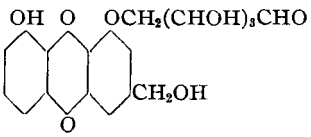
The anthraquinone-glucosidal nature of aloin is shown by the splitting off of aloe-emodin (Tschirch (16), Oesterle (17)) and by its reaction with nitric acid. Aloe upon treatment with nitric acid gives chrysamic acid (Schunck (18)); aloin was shown to be the reacting substance (Stenhouse (19)). Chrysamic acid is to be thought of as a tetranitrochrysazin (Liebermann and Giesel (20)).



By the oxidation of aloin is formed alochrysin ($C_{15}H_8O_5$ Oesterle (17)). The oxidation with Caro's acid liberates tri- and tetraoxymethylanthraquinone (Seel (21) & Scharf (22)). The chromic acid oxidation liberates rhein (Oesterle (17)).

By long standing of an alcoholic solution (Leger), or by the action of alcoholic hydrochloric acid (Oesterle (17)), or by the passage of air through an alkaline solution of aloin (Tschirch and Pedersen (23)), or by the action of sodium peroxide (Seel (21)), aloe-emodin is split off. Barbaloin treated with sodium peroxide liberates a pentose. By treatment of aloin with hydrogen chloride in alcoholic solution, a crystalline decomposition product and a sugar are formed (Rochleder and Czumpelik (24)).

Aloin is a pentoside which upon hydrolysis yields aloe-emodin and *d*-arabinose.



This formula, $C_{20}H_{18}O_9$, was promulgated by Leger and is the one now generally accepted.

In aloe, aloin is associated with other related substances. An isomer, isobarbaloin, $C_{14}H_5O_2$ (CH_2OH). OH-O(8)-$CH_2(CHOH)_3$ CHO, is found in Barbadoes and Curacao aloes; in large amounts in Jafferabad aloe; in very small amounts in Socotrine aloe; and not at all in Cape and Uganda aloes.

Another companion compound of Barbaloin is the non-crystallizable *B*-barbaloin, an optical isomer of barbaloin, from which Leger made a crystalline tetrachloride, $C_{21}H_{16}Cl_4O_9$-5 H_2O. A third, aloesol, was recognized by Leger as a new phenol, and forms another crystalline tetrachloride, $C_{11}H_4Cl_4O_3$. It occurs in Cape and Uganda aloes, in traces in Jafferabad aloe, and not in Barbadoes and Curacao aloe. Aweng (25) obtained a "soluble oxyanthraquinone glucoside" from Barbadoes aloe and Hoffbauer from Curacao aloe. Tschirch and Hoffbauer (26) state that other substances are present which yield chrysamic acid.

Aloe-emodin occurs as a decomposition product of aloin.

The resinous material consists chiefly of a resinotannol ester of cinnamic acid

(Curacao and Barbadoes aloes) or of a resinotannol ester of paracumaric acid (Cape aloe).

A pale yellow volatile oil may be obtained by distillation with 1% potassium hydroxide solution. The oil imparts the characteristic odors to the different varieties and its composition varies.

QUANTITATIVE METHODS FOR THE EVALUATION OF ALOE.

For many years investigators have worked upon methods for the evaluation of aloe. The methods which they recommend give widely divergent results when applied to any one variety of aloe; therefore, they appear not to be accurate quantitative chemical determinations of the aloin present. For these reasons a study was made of these analytical methods with the hope that such a study might contribute to the formulation of a chemical process for the determination of aloin in aloe.

The amounts of moisture and ash present indicate the age of the drug and also the care with which it has been prepared. The U. S. P. X requires that aloe should yield not more than 4 per cent of ash nor more than 10 per cent of moisture. These tests were made in the prescribed manner and the following results were obtained:

	Moisture.		Ash.	
Barbadoes	5.09	5.28%	1.63	1.55%
Cape	5.31	5.31%	0.817	0.724%
Socotrine	3.70	3.89%	2.46	2.47%

The following method described by Tilden (27) is regarded by H. C. Plenge (28) as the best practicable plan of preparing aloin on the small scale from most varieties of aloes:

> Twenty-five grams of the sample should be dissolved in boiling water, the liquid acidified with hydrochloric acid, and allowed to cool. It is then decanted from the precipitated resinous matter, evaporated to about 50 cc., and set aside for two weeks for crystals to form. The liquid portion is then poured off, and the crystals pressed between folds of filter paper. The crude aloin thus obtained is contaminated with a considerable quantity of resin, from which it is best purified by treatment with ethyl acetate, with occasional agitation, till the liquid acquires a brown color, and the yellowish color of the crystals can be distinguished. The liquid is then quickly and carefully poured off, and the crystals dried. Treated in this manner, Barbadoes aloes, for which the method is specially adapted, gave an average of 9% of aloin; whereas Curacao averaged 7.5% and Bonare 7%. Socotrine aloes yielded 3%, but on repeating the process on the same sample, no aloin could be obtained. However, it was isolated to the amount of 10% by digesting 2 parts of the aloes in 3 parts of alcohol for 24 hours, and then heating the liquid over a water-bath for 2 hours. After cooling, the liquid was poured off from the resin, filtered, and set aside in a loosely covered dish to crystallize. The crystals of aloin were washed with a little alcohol and dried.

Tilden's method was applied to three varieties of aloe and these results were noted:

Barbadoes	17.24–16.88%
Cape	4.04– 5.32%
Socotrine	9.04–11.72%

The method may well be used for the laboratory preparation of aloin, but as a quantitative determination it does not appear to be accurate because all of the aloin present probably does not precipitate, and the contaminating resin is not easily completely removed.

For the estimation of aloin in aloes, Schäfer (29) has proposed the following method, based on the fact that aloin, in ammoniacal solution, forms compounds with the alkaline earths, which are but slightly soluble, and from which the aloin can be recovered on treatment with an acid:

Fifty grams of aloes are treated with 300 cc. of boiling water containing a few drops of hydrochloric acid, and when cold the solution is separated from the resin. Fifty cc. of 20% ammonia and 30 cc. of 50% calcium chloride are then added, and the whole well shaken. After 15 minutes, the precipitate is separated, and after being pressed is triturated in a mortar with a slight excess of hydrochloric acid. The aloin and calcium chloride are dissolved in as little boiling water as possible and filtered. On cooling, the aloin separates in crystals. By this method, Schäfer found from 15 to 30% of aloin in aloes of different origin.

This method yielded:

Barbadoes	25.48–29.34%
Cape	15.30–17.00%
Socotrine	12.74–13.02%

The precipitate appeared to include an appreciable amount of resin which would tend to make the results high.

Leger's (30) method is as follows:

Five hundred grams of the sample are boiled with a mixture of 1800 cc. of chloroform and 600 cc. of methanol for four hours under a reflux condenser. After settling, the supernatant liquid is decanted and distilled, and the residue taken up in absolute alcohol, from which crystals of aloin separate in 3 or 4 days. By this means, 5 to 6% of aloin was obtained from Cape aloes and from Barbadoes aloes, 10% from Curacao aloes and as much as 20% from Jafferabad aloes. Socotrine aloes gave only 4%. The method as described above cannot be considered as possessing the attributes of a quantitative method, but Leger's object was mainly to get large quantities of the aloins to serve for their separate detection and approximate estimation.

Tschirch and Hoffbauer (26) propose the determination of the active ingredients as the part soluble in a methanol chloroform mixture. The active ingredients include, not only aloin, but aloe-emodin and also other materials which have purgative properties. The residue is regarded as worthless resin.

Five grams of aloe are macerated for 2 hours in a 50-cc. flask with 5 cc. of methanol. The liquid is then heated to 50–60°, 30 cc. of chloroform added gradually with agitation, and the whole allowed to stand for half an hour. The yellow-colored liquid is filtered from the separated resin through a small fluted filter into a weighed Erlenmeyer flask and the methanol-chloroform mixture distilled off and used again to extract the material in the first flask. The second methanol-chloroform extract is filtered into the tared flask, and the solvent again distilled off and used to extract the residue in the first flask. This procedure is repeated four times. According to Tschirch and Hoffbauer, the extracted material, when dried at 100°, should weigh not less than 4 Gm. 81 to 87% active material was obtained from Cape aloe, 67% from Curacao, and 37% from Socotrine aloe.

The procedure was applied to the same three varieties with the following results:

Barbadoes	56.34–57.42%
Cape	75.28–77.73%
Socotrine	49.26–49.35%

These results were lower in the case of Barbadoes and Cape aloes and higher in the case of Socotrine aloe. Some difficulty was encountered in obtaining concordant results.

L. v. Itallie (31) considers the above method open to objection on the ground that much material remains in the original flask and is included as resin which is not resin at all, but other substances protected from the solvent action of the chloroform by a coating of resin. He recommends the following modification:

Five grams of the powdered sample are warmed with 5 cc. of methyl alcohol in a 50-cc. flask until a homogeneous liquid is obtained. Thirty cc. of chloroform are then added, and the mixture violently and continuously shaken for 5 minutes. By this time most of the separated resin will have adhered to the flask. After standing till clear, the liquid is decanted. The residue is again dissolved in methyl alcohol, and the resin again precipitated by means of chloroform, and this treatment is repeated once more.

In this way L. v. Itallie found 57 to 82% active material in Cape aloe and 79 to 89% in Curacao aloe. This method yielded:

Barbadoes	76.48–76.92%
Cape	82.47–82.84%
Socotrine	70.31–70.39%

Although L. v. Itallie objects to the method of Tschirch and Hoffbauer, it might be said with equal weight that the use of three separate portions of methanol extracts considerable resin and the result is higher than it should be.

The same author attempts the determination of aloin as tribromaloin. With pure aloin the method was successful but, applied to aloes from the Cape and Curacao, it indicated over 70% of aloin, which L. v. Itallie considers to be much too high.

Tschirch & Hoffbauer (26) have estimated aloin on the basis of the green fluorescence produced when borax is added to a solution of aloe.

The material extracted by the methanol-chloroform mixture is dissolved in hot water without filtering. Four hundred cc. of a saturated borax solution is added to make about one liter. The solution is then diluted until the fluorescence is barely perceptible. This fluorescence is shown by barbaloin diluted 1 to 250,000. Their results indicated the presence of 16–20% of aloin in Cape aloe, 16–18% in Barbadoes, 18% in Curacao, and 8% in Socotrine aloe. Applied to 3 varieties of aloe, this method showed:

Barbadoes	12.96–12.99%
Cape	16.23–17.15%
Socotrine	11.14–12.67%

Little difference is shown in the strength of the different varieties. The end-point is not distinct, and the fluorescence fades in a short time. The test must be completed in as little time as possible.

A consideration of the constitution and configuration of aloin suggested a possible method for its quantitative analysis, namely, the hydrolysis of the aloin with the splitting off of a pentose (arabinose), the conversion of the pentose to furfural and the precipitation of the furfural by means of phloroglucinol.

METHOD OF PROCEDURE.

The phloroglucinol (symmetrical trihydroxybenzene) solution is prepared by dissolving 11 Gm. of phloroglucinol in 300 cc. of warm 12% hydrochloric acid. This solution is poured into enough cold 12% hydrochloric acid to make 1500 cc., and the liquid allowed to stand several days for crystallization of diresorcinol and filtered immediately before using.

Kröber (32) proposed the following method for pentoses.

A quantity of the material, chosen so that the weight of the phloroglucide obtained shall not exceed 0.300 Gm., is placed in a flask with 100 cc. of 12% hydrochloric acid and several pieces of recently heated pumice stone. The flask is placed on a wire gauze, connected with a condenser and heated, rather gently at first, so that 30 cc. are distilled over in about ten minutes, the distillate passing through a small filter paper. The 30 cc. distilled are replaced by a like quantity of 12% hydrochloric acid which is added by means of a separatory funnel in such a manner as to wash down the particles adhering to the sides of the flask, and the process continued until the distillate amounts to 360 cc. To the completed distillate is added gradually a quantity of the phloroglucinol solution and the mixture stirred thoroughly. The amount of phloroglucinol used should be about double that of the furfural expected. After the addition of the phloroglucinol, the solution first turns yellow, then green and very soon an amorphous greenish precipitate appears, which grows rapidly darker until it finally becomes almost black. The solution is made up to 400 cc. with 12% hydrochloric acid and allowed to stand over night.

The amorphous black precipitate is filtered through a tared Gooch crucible and washed carefully with 150 cc. of water in such a way that the water is not entirely removed from the crucible until the very last. The crucible is then dried for four hours at 100° and weighed. The increase in weight is calculated as phloroglucide.

a For weight of phloroglucide "a" under 0.03 Gm.: pentoses = $(a + 0.0052) - 1.0170$.

b For weight of phloroglucide between 0.03 Gm. and 0.30 Gm.: Kröber's table is used.

Kröber's method was applied to commercial aloin and also to Barbadoes aloe with the following results:

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	5.4344 Gm.	0.0207	0.0263	0.0048
Barbadoes aloe	6.7921	0.0214	0.0271	0.0040
Barbadoes aloe	10.1710	0.0287	0.0345	0.0034

The amount of pentose shown here is very small. It may occur free in the aloin or aloe or it may be formed by hydrolysis during the process.

Various methods were tried in the attempt to hydrolize aloin. Commercial aloin and Barbadoes aloe were refluxed separately with 3% hydrochloric acid for four hours, the acid content was raised to 12% and the amount of pentose determined as above.

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	6.4136 Gm.	0.0244	0.0301	0.0047
Barbadoes aloe	5.7726	0.0214	0.0271	0.0047
Barbadoes aloe	10.4554	0.0318	0.0411	0.0040

The amount of pentose found is approximately the same as in the case where no preliminary heating with acid was done, and it may be concluded that no hydrolysis of aloin takes place under such conditions.

Commercial aloin was placed in a flask and 100 cc. of alcohol containing 3% hydrochloric acid were added. The flask was connected to a reflux condenser and the mixture refluxed by heating on a water-bath for four hours. The alcohol was distilled off, 100 cc. of 12% hydrochloric acid added and pentoses determined. The process was repeated with Barbadoes aloe.

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	1.0179	0.0085	0.0139	0.0134
	1.1351	0.0098	0.0152	0.0134
Barbadoes aloe	3.7751	0.0363	0.0460	0.0122
	4.2682	0.0396	0.0497	0.0116

The amount of pentose found is still very small and indicates only incomplete hydrolysis of the aloin.

Commercial aloin was placed in a flask and 100 cc. of 3% potassium hydroxide solution added. The flask was connected to a reflux condenser and heated for four hours. The potassium hydroxide was neutralized with concentrated hydrochloric acid and enough hydrochloric acid added in addition to bring the acid content to 12%. Arabinose was then determined. The process was repeated with Barbadoes aloe.

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	0.9543	0.0068	0.0122	0.0128
	0.9891	0.0073	0.0127	0.0128
Barbadoes aloe	4.0475	0.0210	0.0266	0.0066

The results obtained here are approximately the same as those in which 3% hydrochloric acid in alcohol was used.

Commercial aloin was placed in a flask and 100 cc. of alcohol containing 3% potassium hydroxide added. The flask was connected to a reflux condenser and heated over a water-bath for four hours. The potassium hydroxide was neutralized with hydrochloric acid and the alcohol distilled off. One hundred cc. of 12% hydrochloric acid were added and the arabinose determined. The same procedure was repeated with Barbadoes aloe.

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	0.9853	0.0146	0.0201	0.0205
	0.9647	0.0176	0.0232	0.0240
Barbadoes aloe	3.7711	0.0328	0.0421	0.0112

If the molecular weight of aloin is taken as 402 and that of arabinose as 150, the equivalent for converting arabinose to aloin becomes 2.680. The per cent aloin hydrolyzed is found by multiplying the weight of arabinose per Gm. of substance used by 2.68×100. In the first determination, $0.0205 \text{ Gm.} \times 2.68 \times 100 = 5.49$ per cent; in the second determination, $0.0240 \text{ Gm.} \times 2.68 \times 100 = 6.43$ per cent. The hydrolysis of the aloin was not complete by this method.

Mild oxidizing agents were next used to split off the pentose. Commercial aloin was dissolved in 75 cc. of water and the solution boiled. Sodium peroxide was added in small portions, the alkali formed neutralized with hydrochloric acid and the acid content raised to 12%. The pentose was then determined. This procedure was repeated with Barbadoes aloe.

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	0.8275	0.0240	0.0297	0.0358
	0.8756	0.0220	0.0277	0.0317
Barbadoes aloe	2.4422	0.0134	0.0169	0.0078
	1.8526	0.0111	0.0166	0.0089

In the case of commercial aloin, 0.0356 Gm. pentose per Gm. material taken is equivalent to 9.59% aloin; 0.0317 Gm. pentose is equivalent to 8.50% aloin.

Commercial aloin was dissolved in 75 cc. of water and the solution boiled. Five grams of sodium perborate were added in divided portions to the boiling liquid. The alkali formed was neutralized with hydrochloric acid, the acid content brought to 12% and the pentose determined. The method was repeated with Barbadoes aloe.

Substance.	Wt. Taken.	Phloroglucide.	Pentose.	Pentose per Gm.
Commercial aloin	0.8795	0.0904	0.1055	0.1195
	0.8788	0.0760	0.0897	0.1021
Barbadoes aloe	2.7340	0.0674	0.0802	0.0293
	3.4388	0.0726	0.0860	0.0250

In the case of commercial aloin, 0.1195 Gm. arabinose is equivalent to 32.0% aloin; 0.1021 Gm. arabinose is equivalent to 27.4% aloin. For Barbadoes aloe, 0.0293 Gm. arabinose indicates 7.8% aloin; 0.0250 Gm. indicates 6.7%. These results are not sufficiently high to warrant the use of the method as a quantitative determination of the aloin present.

SUMMARY.

1. A laboratory review of the methods proposed by previous investigators shows that, at present, there is no satisfactory method for the evaluation of aloe or for the quantitative determination of the aloin present.

2. The configuration of aloin suggests a method for its determination, namely, by hydrolizing the aloin so as to split off aloe-emodin and arabinose (a five-carbon-atom sugar). The arabinose is then converted to furfural which is precipitated by means of phloroglucinol.

3. The attempted hydrolysis of aloin with 3% hydrochloric acid in water showed no appreciable hydrolysis.

4. The use of 3% hydrochloric acid in alcohol in the attempt to hydrolize aloin showed only slight hydrolysis.

5. The attempted hydrolysis of aloin with 3% potassium hydroxide in water gave no better results.

6. The hydrolysis attempted with 3% potassium hydroxide in alcohol was very incomplete.

7. Treatment with sodium peroxide indicated the splitting off of arabinose from approximately 9% of the aloin.

8. Treatment with sodium perborate indicated the decomposition of approximately 30% of the aloin.

9. The work of Grönewold (10), who prepared a tri- and a hexa-acetyl derivative, suggests the possibility of determining aloin by acetylization, whereby acetyl groups replace the hydroxyl groups found in the aloin molecule. The precipitated acetyl derivative would be washed free from acetic anhydride, the acetyl groups liberated as potassium acetate by refluxing with alcoholic potassium hydroxide. Excess sulphuric acid would be added and the acetic acid distilled into a volumetric sodium hydroxide solution and the excess titrated with sulphuric acid. The amount of sodium hydroxide neutralized indicates the number of acetyl groups introduced. From preliminary work on this method, it would appear to be worthy of further study.

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CHEMICAL AND PHYSICAL DETERMINATIONS ON THE GUM AND VOLATILE OIL OF ASAFŒTIDA.

BY JOSEPH F. CLEVENGER.*

It has been apparent for some time that the method of assay of asafœtida and the requirements for that product, as given in the U. S. P. X, are not entirely satisfactory. The New York Station has recently made a study of the subject and as a result has adopted a method of assay which gives figures that appear to be a truer measure of the quality of the drug.

To obtain information to serve as a basis for suggesting a suitable quality standard for asafœtida, each of 41 lots of the gum offered for entry at the port of New York during the past three years was analyzed. Samples were made uniform by passing them through a meat grinder and for the determinations of alcohol-soluble extractive, acid-soluble ash, moisture content and yield of volatile oil the following methods were adopted:

ALCOHOL-SOLUBLE EXTRACTIVE.

Weigh 10 Gm. of the sample in a tared 250-cc. Erlenmeyer flask. Add 100 cc. of alcohol. Attach a reflux condenser and boil for one hour or until the sample is completely disintegrated. Dry a filter paper in an oven at 110° C., cool and weigh. Fit the paper in a Buchner funnel of slightly smaller diameter so that the edges turn up approximately one-fourth of an inch. Filter the gum and alcohol hot, under suction, taking care not to fill the Buchner funnel above the edges of the filter paper. Wash with hot alcohol until no cloudiness is produced when the filtrate is

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