group produces a rise in boiling point of about 16°. The effect of the density is illustrated graphically in the accompanying figure. The decrease in the density of the methoxymethyl salts with rising molecular weight is analogous to the behavior of the chloro ethers, investigated by Favre.[1] All of the liquids freeze below —79°. While the two lower compounds are very unstable in the presence of water, none of the liquids fume in air.

Two of the present authors are engaged on further studies of these and similar reactions. We desire to thank E. Emmet Reid for important suggestions.

MORGANTOWN, W. VA.

[CONTRIBUTION FROM THE LABORATORY OF ORGANIC ANALYSIS OF THE UNIVERSITY OF ILLINOIS.]

THE QUALITATIVE IDENTIFICATION OF THE DRUGS CONTAINING EMODIN.[2]

BY GEORGE D. BEAL AND RUTH OKEY.

Received February 27, 1917.

Cascara, Rumex, Rhubarb, Frangula, Senna, and Aloes are the best known members of the group of drugs which are supposed to owe their medicinal properties to the fact that they all contain some derivative of a methyl anthraquinone, such as emodin, chrysophanic acid, or their homologs or isomers, or glucosides yielding these compounds on hydrolysis. A summary of the most authentic analyses of these drugs shows the following compositions:

Alexandrian Senna.[3]—The anthraquinone derivatives are rhein, aloe-emodin and their glucosides. In addition to these are the other substances commonly found in plant drugs, such as sugars, alcohols, acids, and resins.

Cascara Sagrada[4] contains emodin and iso-emodin, but no glucosides of emodin or chrysophanic acid. In addition to the other common substances are found syringic acid and the alcohol rhamnol.

Rumex Ecklonianus.[5]—This drug contains, in the overground portion of the plant, emodin, emodin-mono-methyl ether, and chrysophanic acid.

Rumex Crispus.[6]—No recent analysis of this drug could be found.

Rhubarb.[7]—Contains rhein, emodin, emodin-mono-methyl-ether, aloe-emodin, chrysophanic acid and their glucosides, as well as a new anthraquinone, rheinolic acid.

[1] Favre, *Bull. soc. chim.*, [3] 11, 879 (1894).

[2] Presented in part at the Urbana meeting of the American Chemical Society.

[3] Tutin, *J. Chem. Soc.*, 103, 2006 (1913).

[4] Jowett, *Proc. Am. Pharm. Assoc.*, 52, 228 (1904).

[5] Tutin and Clewer, *J. Chem. Soc.*, 97, 1 (1910).

[6] The authors are now engaged in a study of its proximate composition.

[7] Tutin and Clewer, *J. Chem. Soc.*, 99, 946 (1911).

Rhamnus Frangula.—No recent or complete analyses of frangula have been published. The presence of emodin and its glucoside frangulin and of rhein and chrysophanic acid seems to be established.

Aloes.[1]—The anthraquinone derivatives are aloe-emodin and its glucosides. There are also a large number of wax alcohols, fatty acids and resins present.

Since the best qualitative tests for these drugs probably depend on certain properties of the anthraquinone compounds present, it seems well at this point to devote a little space to considering their characteristics.

Aloe-emodin from the nature of its reduction products, seems to be a derivative of β-methyl anthracene. It is obtained from the ether extract of the drug and crystallizes in orange-red needles, melting at 224°. Ether, hot alcohol, benzene, ammonia, and the fixed alkalies dissolve it, the last two solutions having a deep red color.

The principal derivative is chrysammic acid, obtained on heating with nitric acid. This forms yellow plates or monoclinic prisms, and is soluble in alcohol and ether, but almost insoluble in water.

Chrysophanic Acid on reduction with zinc dust yields β-methyl anthracene. It is also obtained from the ether and petroleum ether extracts of the drugs containing it, usually closely associated with emodin-mono-methyl-ether. It forms golden-yellow plates melting at 198° when pure, and is soluble in water and the cold solutions of the alkali carbonates, but soluble in dilute solutions of the alkali hydroxides; rather sparingly soluble in ether and alcohol, but quite soluble in benzene and chloroform.

By heating with strong ammonia, or even on long standing with this reagent, it forms mono-amino chrysophanic acid; and on warming with concentrated nitric acid a tetranitro derivative is formed.

Rhein may be obtained by extracting the drug resins containing it with dilute solutions of alkali carbonates, or by oxidizing aloe-emodin with chromic acid. It forms small yellow needles melting at 321°, and is of a distinctly acid character, slightly soluble in organic solvents, but forming solutions of the characteristic red color with concentrated sulfuric acid, ammonia, and the fixed alkali carbonates and hydroxides.

Emodin on reduction with zinc dust yields β-methyl anthracene. It may be extracted from the plant by alcohol, and separated from the ether solution of the resin so obtained by extraction with sodium carbonate. It crystallizes from glacial acetic acid in orange-red needles; and is soluble in ammonia, the alkali carbonates and hydroxides and concentrated sulfuric acid with a red color.

Emodin-mono-methyl-ether has practically the same solubilities as chrysophanic acid, with which it is closely associated.

[1] Tutin and Naunton, *Pharm. Jour.*, **91**, 836 (1914).

Glucosides.

Aloin, on hydrolysis, yields aloe-emodin and a substance as yet undetermined. It is only slightly soluble in cold water, but more soluble in hot water and alcohol, while it is insoluble in the other ordinary organic solvents. It dissolves in the alkali carbonates and hydroxides with an orange color, and these solutions show a yellow-green fluorescence.

Frangulin, on hydrolysis, yields emodin and rhamnose. It is insoluble in ether and water, but soluble in benzene and hot alcohol. Sulfuric acid and the fixed alkalies dissolve it with a red color.

Buchner[1] mentioned the first qualitative test for these drugs—the purple-red color with alkalies. Dragendorff[2] gave a series of tests for aloes with lead acetate, platinic chloride, gold chloride, and mercurous nitrate, which last reagent we have used. More attention has been paid to aloes than to the other drugs and it would be possible to cite a large number of papers. Lenz[3] published an account of the nitric acid-potassium cyanide test for aloes, which we have used for group identification.

The most recent work is that of Bailey[4] in which he discusses the nature and reaction of the anthraquinone fractions obtained from the different drugs. He does not, however, offer a qualitative scheme for the identification of all the drugs.

The object of our experiments was to find a simple system of tests by which the preparations of these drugs might be distinguished, one from another, or by which they might be determined in mixtures. These tests were to be based, if possible, on the behavior of the anthraquinone derivatives present, and especially on the formation of oxidation and reduction products which differed in color.

Preparation of Solutions.

The extracts of the drugs used in testing were, as a rule, made by shaking one part of the fluidextract, or of a solution made by extracting a solid drug preparation with boiling 50% alcohol with four parts of an immiscible solvent, allowing the mixture to settle, and drawing off the solvent for use. Ethyl ether, benzene, and amyl alcohol proved most satisfactory for this purpose.[5]

Tests Applied Directly to the Solutions in these Solvents.

Ammonia Test.—On shaking the benzene solution with an equal vol-

[1] *Neues Repert für die Pharm.*, **2**, 145 (1853).

[2] "Beitrage zur gerichtlicher Chemie einzelner Gifte."

[3] *Z. anal. Chem.*, **21**, 220 (1883).

[4] *Am. J. Pharm.*, **87**, 145 (1915).

[5] The colors obtained in these tests were named according to the color charts of the Milton Bradley Company, found in Mulliken's "Identification of Pure Organic Compounds." (John Wiley & Sons, New York.) Throughout this paper we have given the shades of color obtained with the concentrations recommended for the test solutions. This must be remembered in attempting to apply this system of analysis.

ume of concentrated ammonia, a violet-red precipitate (VR, tint 2), which gradually collects between the benzene and the aqueous layers, is obtained with rhubarb. Except when phenolphthalein is present,[1] the red colors which are developed here in the ammonia layer by the anthraquinone drugs are accepted as a general test for the group. The precipitate from rhubarb may be due to the formation of an aminoanthraquinone derivative. Since other drug resins which might give a precipitate under the same conditions can be distinguished by the fact that these precipitates will be of a brownish color, this is a very good test for rhubarb in mixtures.

Shaking the amyl alcohol solutions of cascara and aloes with ammonia produces a brilliant green-red fluorescence, while the other drugs show only the characteristic violet-red color of the group. It was found, however, that this test failed with old fluidextract of cascara, while age seemed to have little or no effect on the aloes. There is no mention of the use of this test in the literature, although it is well known that solutions of certain anthraquinone glucosides, such as aloin, are fluorescent.

Mercurous Nitrate Test.—The amyl alcohol solution of aloes, when shaken with a saturated solution of mercurous nitrate, containing a slight excess of nitric acid develops a brilliant dark red color in the aqueous layer. Usually from two to eight minutes are required for this test to reach its maximum, after which the color fades to a reddish brown. If a very small amount of aloes is present in a mixture, the color will be pink (OR, tint 2, or R, tint 2) and will disappear sooner. The solutions of the other drugs remain colorless throughout the test. Dragendorff[2] mentioned the fact that Barbadoes and Curaçao aloes were colored reddish by mercurous nitrate, but he described no method of applying the test; and there is no mention of it in the subsequent literature.

Nickel Acetate Test.—The ether extracts, on shaking with a saturated solution of nickel acetate, show striking variations in color. Senna develops a dark brilliant red (R, shade 1) immediately, becoming violet (VR) on the addition of a small amount of 10% KOH solution. Rhubarb and frangula require the addition of alkali to produce the color change (VR, shade 1); and it is noticeable that the color from rhubarb is much more soluble in ether than that from the other drugs. Cascara treated in this way is orange-yellow before (Y, shade 1) and dark red (R, shade 2) after adding the alkali. Rumex, if the ether solution is kept well mixed with the acetate, develops no color, retaining the green of the nickel salt throughout the test. Aloes is yellow-brown with these reagents. These red colors with nickel acetate have been mentioned before as a reaction

[1] For methods of distinguishing this drug, see the paragraph on Group Identification.

[2] *Loc. cit.*

of emodin, but they have apparently not been used as a means of distinguishing the drugs containing it.

Lead Subacetate Tests.—The amyl alcohol solution of aloes, when shaken with lead subacetate, became red (R), while the other drugs did not change color. The color test with this reagent has not been used, although there is mention of several precipitation tests with the subacetate.

The benzene solution of rhubarb, when shaken with lead subacetate, gave a yellow-orange precipitate (YO, shade 1) which turned red with alkali, while the precipitates from the other drugs remained white. It was found that this test served to detect rhubarb in mixtures where the effectiveness of some of the other tests was spoiled by the highly colored or resinous extracts of other substances.

The **iodine test** which has been given for amyl alcohol and benzene solutions of these drugs is valueless, because the color may be obtained with the reagents alone.

The **borax test** for aloes, given by Schoutelen,[1] which depends on the fluorescence of dilute aqueous solutions of the drug with this reagent, served fairly well for aloes alone, but was spoiled by the resinous character of some of the other drugs.

Tests Applied to the Residues Obtained by Evaporating the Solutions Described Above.[2]

Nitration Tests.—The films were moistened with concentrated nitric acid and again evaporated to dryness. The colors of these residues varied from yellow to red-brown. Rumex and senna were always lighter in color than the others; and the solution of aloes in benzene gave a light yellow residue, while that from the other solvents was a deep red.

When these nitrated residues were covered with saturated stannous chloride solution containing a little free HCl, those from the ether and benzene solutions of senna turned green (GY, med) those from aloes, reddish brown (RO, shade 2, dark) and the others varying shades of red-violet, that from rumex being the lightest, and frangula the deepest. It was found advantageous in applying this test, especially with mixtures, to decant the stannous chloride solution, wash the residue, which will adhere to the dish, with a jet of distilled water, decant again, and treat with alcohol. The residues from the cascara and frangula solutions are the most soluble in alcohol, the cascara being a deep red (med R); the frangula a brilliant purple (RV, shade 1); rhubarb a deep violet (RV); rumex, light violet (VR, tint 1); while aloes is dark yellow-brown (O, shade 2); and senna is green (YG shade 1). The green residue from the senna solution becomes red-purple on the addition of alkalies. and light red on

[1] *Z. anal. Chem.*, 31, 723 (1892).

[2] These were prepared in thin films by evaporating 1–2 cc. portions of the various extracts on small, white porcelain dishes, using a steam bath.

treatment with U. S. P. solution of chlorinated soda, while the residues from the other drugs become brown before decolorizing. This color does not appear with chlorine water alone, and it is necessary that the chlorinated soda solution have exactly the strength indicated in the Pharmacopeia.

On repeating these tests with ferrous sulfate instead of the stannous chloride, it was found that the colors of the iron salts masked any others. No other reducing agents were found to be successful.

Attempts were made to take advantage of the diazo reaction by treating the stannous chloride reduction products with acid solutions of sodium nitrite in ice-water. On the addition of alkali to this mixture senna, rhubarb, and frangula gave red tints, cascara, violet-red, and rumex a light red-violet, while aloes was yellow-orange. While, with careful manipulation, the colors were distinct enough with the individual drugs, they did not give good results with mixtures.

By coupling the diazotized residues with various organic bases, a number of very brilliant reds and greens were obtained. These were, however, too much alike to be of service in the solution of our problem.

More completely nitrated products were prepared by the use of a mixture of sulfuric and nitric acids at the temperature of the steam bath. The intensity of these colors was increased by partial neutralization with sodium and barium carbonates, cascara and frangula becoming a deeper and darker red-orange, rhubarb only a little less deep, and the colors of the other drugs varying from yellow to orange.

The effect of chlorine water and of sodium hypochlorite solutions in oxidizing the residues from the stannous chloride reductions was determined with results that were unsatisfactory from an analytical point of view, save in the case of the senna reaction mentioned above.

Tests Applied to the Resins Precipitated by Adding the Concentrated Alcoholic Extracts of the Drugs to an Excess of Water.

Sodium Peroxide Test.—Alvarez[1] stated that if emodin were treated in a porcelain crucible with alcohol and sodium peroxide, followed after a short time by the addition of water, a red color would be developed in the solution. Using this method, we tested the dried resins precipitated by pouring the fluidextracts of these drugs into a large excess of water. Rhubarb gave a red color with the alcohol alone, and an orange-red in the aqueous solution. The other drugs showed no colors in the alcoholic, and varied as follows in the water solutions: senna, yellow-orange; cascara and rumex, orange; aloes, orange-yellow; and frangula, orange-red. This suggests interesting possibilities in the anthraquinone content of rhubarb.

[1] *Ann. chim. anal.*, 12, 9; *J. Chem. Soc.*, **92**, 143 (1907).

Summary.[1]

Tests for Senna.—The ether solution, shaken with a saturated solution of nickel acetate, shows a deep red color, while the other drugs vary from green to deep orange-brown. With potassium hydroxide a violet precipitate is formed, the ether returning to the original color.

If the ether residue is evaporated with concentrated nitric acid, on covering with a stannous chloride solution, senna will become green, aloes brown, and the other drugs violet. The benzene solution of senna gives the same reaction, but the amyl alcohol solution is less satisfactory. The green reduction product from senna turns bright red when first treated with U. S. P. solution of chlorinated soda, while the products from the other drugs turn yellow or brown.

The nitrated, reduced, and diazotized residues from senna are red, while those from the other drugs are tints of red-violet. The red color is harder to destroy with chlorine water than those from the other drugs.

Tests for Cascara.—The amyl alcohol solution from a reasonably fresh fluidextract of cascara, made after the extraction with benzene, shows a green fluorescence with concentrated ammonia. Aloes also responds positively to this test, but may be differentiated in other ways. If aloes is present, cascara may be identified by nitrating, and reducing with stannous chloride. The aloes residue will be yellow, and that from cascara a deep red.

Cascara, frangula, and to a less extent, rhubarb residues from the benzene extract, nitrated with the aid of sulfuric acid, and then not quite neutralized with a carbonate, give an orange-red color, while the other drugs develop little more color than that of the nitrated solutions.

Tests for Rumex.—(R. crispus) when thoroughly mixed with nickel acetate solution, rumex is the only drug which retains the color of the nickel salt, even after the addition of alkali.

By nitrating with the aid of sulfuric acid, half neutralizing, and reducing with stannous chloride, rumex gives a light red color, while aloes after this treatment is orange-yellow, and senna yellow.

Tests for Rhubarb.—Shaking the benzene extract with strong ammonia gives a red-violet precipitate settling between the two liquids. The other drugs give the deep red color, but no precipitate. Rhubarb also gives a characteristically colored precipitate with lead subacetate.

The ether solution, on shaking with an equal amount of a saturated solution of nickel acetate and then adding 10% KOH, gives a dark red-violet precipitate like that from frangula, but the ether layer from rhubarb becomes an intense red, retaining its color until after the ether solutions of all the other drugs have become colorless.

[1] For details of manipulation, and exact shades, see description above.

The resin precipitated by pouring the fluidextract into water, after thorough drying, becomes red immediately upon treatment with sodium peroxide and 95% alcohol, while no color is developed with the other drugs until after the addition of water.

Tests for Frangula.—Frangula gives, when the ether solution is treated with nickel acetate and potassium hydroxide, a deep red-violet precipitate in the aqueous layer, while the ether layer becomes colorless almost immediately. With rhubarb the ether layer remains colored for a longer time, and the precipitate from cascara is dark orange-red.

Amyl alcohol solutions of cascara and frangula give red colors with nickel acetate and alkali, while the amyl alcohol solutions of senna and rhubarb are red-orange and yellow-orange, respectively, and those from rumex and aloes are yellow to yellow-orange.

By nitrating and reducing the residues from the ether and benzene solutions, a much deeper and brighter violet-red is obtained with frangula than with rhubarb and rumex. Cascara is orange-red, senna green, and aloes brown.

Tests for Aloes.—An amyl alcohol solution of aloes, when treated with mercurous nitrate solution, develops a red color, which lasts for some minutes, becoming brown on long standing.

If the residues from the ether or amyl alcohol extract are taken up with 30% alcohol, to which a drop of dilute copper sulfate solution and a few crystals of sodium chloride are added, a red color is developed. While the other drugs do not respond to this test, we have sometimes found it difficult to get good results when only a small amount of aloes was present in a mixture; and would therefore recommend the mercurous nitrate test. This is the cupraloin test of Klunge.[1]

Solutions of aloes evaporated, and taken up with dilute alcohol, then treated with copper sulfate and hydrogen peroxide solutions, give on boiling a red color which is characteristic of aloes alone.[2]

From the foregoing reactions we have devised the following tentative qualitative scheme for the identification of the different drugs:

Group Identification.

Shake a small amount of a dilute alcohol solution of the drug preparation to be analyzed with four times its volume of benzene. If anthraquinone bodies are present, a small portion of this solution when shaken with 30% NaOH will give a permanent color varying from light red to deep violet. If phenolphthalein is present, a red color will be developed instantly but the concentrated alkali destroys this completely within

[1] *Chem. Ztg.*, 4, 1085 (1880).

[2] Hirschsohn, *Pharm. Zentr.*, 42, 63 (1901).

five minutes, if the test tube is shaken. It has no effect on the colors from the anthraquinone derivatives.[1]

If this test is positive, evaporate another portion of the benzene solution to dryness, moisten with concentrated nitric acid, and evaporate again. The residue will have a red or orange-red color, and when treated with a solution of potassium cyanide in potassium hydroxide (30%) will take on a red or purplish red color in the presence of anthraquinone derivatives. Phenolphthalein gives a brownish color with the cyanide.

Identification of the Individual Drugs.

Divide the solution to be tested into three parts. Shake out one part with four times its volume of benzene, draw off the solvent, and repeat the extraction with amyl alcohol. Make a similar extraction of the second part of the solution, using ethyl ether as the solvent. Reserve the third portion of the sample.

I. Shake a portion of the benzene extract with concentrated ammonia. A deep red-violet color, and a red-violet precipitate settling between the two layers of liquid indicates rhubarb. If a precipitate is formed, but its red-violet color is not distinct, the presence or absence of rhubarb should be confirmed by applying the lead subacetate test, or, if necessary, by preparing a resin and applying the sodium peroxide test.

II. Shake a portion of the amyl alcohol extract with strong ammonia. A deep red color viewed by transmitted light, with a dark green fluorescence, indicates aloes, or a freshly prepared extract of cascara. If this test is positive, shake another portion of this extract with mercurous nitrate solution. If the first reaction was due to aloes, a red color or shade varying with the concentration of the aloes will appear in the aqueous layer. The cupraloin test, the hydrogen peroxide test, and the fluorescence test with borax, may be used to confirm this result.

The presence or absence of cascara may be confirmed, or proven in case the drug was too stale to give the fluorescence test, by evaporating a portion of the benzene or amyl alcohol solution, nitrating, and treating with stannous chloride. Cascara will give a deep red color, aloes a yellow-brown.

III. Shake a portion of the ether extract with an equal volume of saturated nickel acetate solution. A red aqueous layer indicates senna. If the solution retains its green color, and gives a green precipitate with potassium hydroxide, but the preliminary tests have shown a permanent color with this alkali, rumex must be present. This is strictly an elimination test, and is not trustworthy unless the acetate and ether are kept mixed at the time of the addition of the alkali.

[1] For other and more complicated tests for emodin in the presence of phenolphthalein, see Warren, *Am. Jour. Pharm.*, 86, 444–9 (1914), and Bailey, *J. Ind. Eng. Chem.*, 6, 320 (1914).

If, on shaking the above mixture with potassium hydroxide, a violet precipitate is formed, the presence of senna is indicated, while with rhubarb or frangula it will be red-violet, and with cascara dark orange-red. Rhubarb and frangula give orange solutions before adding the alkali. The ether layer, after adding the alkali, will become colorless almost immediately, except in the presence of rhubarb, when the red color persists for a time.

IV. If the above tests have not given conclusive evidence as to the composition of the drug, nitrate a portion of the ether extract, and reduce with stannous chloride, keeping at the temperature of boiling water. Senna gives a green residue, aloes a brown one, cascara, red, and rumex, rhubarb and frangula violet-red, the depth of color increasing in the order named. That from phenolphthalein is lemon-yellow throughout the whole process. Frangula residues are much more soluble in alcohol than the others, and on evaporation leave a much deeper violet ring.

Treat the above residues, after washing with water to remove the excess stannous chloride, with U. S. P solution of chlorinated soda. Senna alone will develop a distinct red color, the others turning yellow before decolorization.

Conclusions.

We have been able to devise color tests which will serve to detect rhubarb, senna, and freshly prepared fluidextract of cascara in almost any proportion in which they would be likely to be found in a commercial mixture.

There were already several good tests for aloes, and we have found a way to apply the mercurous nitrate test.

While we have been able to identify pure extracts of frangula and rumex, and frangula in mixtures in which cascara and rhubarb were absent, the identification of these drugs in other mixtures will require the comparison of the colors obtained in a series of tests.

Phenolphthalein responds to the group reaction with sodium, potassium, and ammonium hydroxides, but when the concentration of fixed alkali exceeds 10% the color disappears rapidly. The colors of the emodin drugs are unaffected by an excess of concentrated alkali.

URBANA, ILL.

[CONTRIBUTION FROM THE BUREAU OF PLANT INDUSTRY, DEPARTMENT OF AGRICULTURE.]

NOTES ON THE RUBBER FROM EUCOMMIA ULMOIDES, OLIVER.[1]

BY ARTHUR F. SIEVERS.

Received February 12, 1917.

Caoutchouc and caoutchouc-like substances occur rather widely in the vegetable kingdom, but it is doubtful whether it occurs in any plant

[1] Published by permission of the Secretary of Agriculture.