A COMPARISON OF ISOPROPYL AND ETHYL ALCOHOLS
AS MENSTRUA IN THE EXTRACTION OF DRUGS

By

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INTRODUCTION

Isopropyl Alcohol, N. F. VIII, ($CH_3CHOH{\cdot}CH_3$), official synonyms - Propanol, Propanol-2, known also as isopropanol, secondary propyl alcohol, and dimethyl carbinol, was first prepared by Berthelot in 1855 by the reaction of propylene with sulfuric acid and subsequent hydrolysis of the sulfuric acid esters (1). At the present time it is a by-product of the petroleum and natural gas industries.

The first use of the compound in the manufacture of Galenics appears to have been in 1922 when Grant and Johns (2) employed it to replace ether and acetone in the preparation of the oleoresins of capsicum, ginger, cubeb, and aspidium, thus reducing the fire hazard as well as the loss by evaporation.

Fuller (3) in 1923 prepared a number of standard preparations of the United States Pharmacopoeia and National Formulary with isopropanol as the solvent instead of ethyl alcohol. He prepared fluidextracts of digitalis, belladonna, cinchona, cascara, and gentian. In all cases the percolation proceeded with the same degree of facility as if the menstruum prescribed had been employed, and the resultant finished products were of excellent appearance with the full content of active principles. He also prepared tinctures of jalap, ginger, aloe, and opium. Those of jalap and ginger contained the full complement of resins that characterize them, and in addition the tincture of ginger was delightfully aromatic and pungent. Tincture of opium conformed to the official standard.

Martindale and Westcott (4) have shown that isopropyl alcohol is safe internally in small doses in diluted form, and should be useful in the manufacture of tinctures. They found that the physical aspects of tinctures made with isopropyl alcohol compared favorably with those made with ethyl alcohol.

In 1938, Marki (5) reported that isopropyl alcohol compared favorably with ethyl alcohol in its extraction capacity for the alkaloidal drugs cinchona and nux vomica but that it gives a somewhat lower yield of extract from rhubarb than does ethyl alcohol. With few exceptions, the preparations made with isopropyl alcohol exhibited a lower stability than those prepared with ethyl alcohol, and considered from the scientific standpoint, the use of isopropyl alcohol embodies no advantage over ethyl alcohol.

Lauwaet (6) has reported that the alkaloid content of extracts of belladonna, aconite, nux vomica, and cinchona prepared with isopropyl alcohol is equal or superior to equivalent preparations made with ethyl alcohol.

In 1944, Chase, Lehman and Crandall (7) prepared tinctures of digitalis using ethyl alcohol and isopropyl alcohol as the menstrua. They reported that isopropyl alcohol dissolves 15 per cent less extractive than ethyl alcohol and yields 50 per cent less ash. Bioassays by the pigeon emesis, the pigeon fatal dose and the U. S. P. cat methods disclosed the isopropyl product to be, respectively, 28.5 per cent more, 11.7 per cent more, and 7.7 per cent less potent than its ethyl alcohol counterpart.

Burlage and Hawkins (1) in 1946 made a study of the following preparations: Nux Vomica Tincture, Belladonna Tincture, Stramonium Tincture, Hyoscyamus Tincture, and Camphorated Opium Tincture. They prepared two tinctures from each drug, one with isopropyl alcohol and the other by the official formula which calls for ethyl alcohol. It was found that isopropyl alcohol compared favorably with ethyl alcohol as a menstruum for Camphorated Opium Tincture and Belladonna Tincture. In Hyoscyamus Tincture, isopropyl alcohol proved to be a better solvent of the total alkaloids. In the case of Nux Vomica Tincture, the tincture containing isopropyl alcohol contained less strychnine when assayed by the Pharmacopoeial method than that containing ethyl alcohol. Isopropyl alcohol was an unsatisfactory menstruum for the preparation of Stramonium Tincture because of the low yield of alkaloids and the high yield of extractive.

Isopropyl alcohol has been used in the preparation of extracts where the solvent is completely evaporated and also in the production of tablet granulations.

Isopropyl alcohol possesses a lower surface tension, a greater fat solvent action, a more rapid killing power on many organisms, and is more economical than ethyl alcohol. It does not produce the exhilaration in the human body that is characteristic of ethyl alcohol, hence there is no object in using it as a beverage, and therefore, it is free from the burdensome government control and taxation to which ethyl alcohol is subjected. In general its effects are very similar to that of ethyl alcohol in regard to absorption, combustion, and elimination. It produces weak irritation of mucous membranes but moderately strong irritation when inhaled. Skin irritation is very slight and similar to that of ethyl alcohol (8). Quantities up to 20 cc., diluted with water have been administered to humans and cause only a sensation of heat and a slight lowering of the blood pressure, but no other noticeable effects.

As shown by the foregoing, some of the studies have been on a qualitative basis, some have not accounted for total extractive, and others have been made without regard for the volume of menstruum required for extraction.

Therefore, this work was undertaken to make a quantitative comparison of ethyl and isopropyl alcohols as menstrua for the extraction of certain drugs.

EXPERIMENTAL

The crude drugs selected for this study were Belladonna Root, Cinchona, Nux Vomica and Ginger. These drugs were selected because they contain different types of active constituents, official assays are available, the active constituents are present in different forms within the drugs, and because they contain other constituents which may affect the ease with which the active constituents are extracted. Belladonna is a root drug and the important active constituent atropine occurs in the form of an ester tropyl tropine. Cinchona is a bark drug which contains a number of alkaloids. It also contains tannin which hinders extraction of the active constituents. Nux Vomica is a seed drug and contains a fat which is an obstacle in the process of extraction. Ginger, a rhizome, contains a volatile oil and resin as the active constituents.

The drugs were extracted with appropriate strengths of ethyl alcohol and corresponding strengths of isopropyl alcohol. In each case, five 200 cc. fractions of percolate were collected in succession. All percolates were examined for their content of active principles, total extractive, and appearance. Total extractive determinations were made because inert extractive material is a frequent cause of subsequent precipitation in liquid Galenics and is sometimes responsible for other forms of deterioration. Also, fat determinations were run on the Nux Vomica percolates.

The crude drugs themselves were assayed by the methods of the United States Pharmacopoeia, XIII, and the National Formulary, VIII, to determine the actual content of active constituents in each drug so that a comparison could then be made with the amount of constituents extracted by ethyl alcohol and isopropyl alcohol in the various percolates.

The detailed procedures are as follows:

1. Belladonna Root

Preparation of Belladonna Root Percolates with Ethyl Alcohol

One thousand grams of the ground drug was mixed with a sufficient quantity of a mixture of four volumes of ethyl alcohol and one volume of water to render it evenly and distinctly damp. This required 700 cc. of solution. The dampened drug was allowed to stand for 15 minutes. It was then packed firmly in a cylindrical percolator and a sufficient quantity of the menstruum (four volumes of ethyl alcohol and one volume of water) was added to saturate the drug and leave a stratum above. When the liquid began to drop from the percolator, the lower orifice was closed, the percolator covered, and the drug allowed to macerate for 48 hours. At the expiration of this maceration period, percolation was conducted at a rate of 1 cc. per minute and five 200 cc. fractions of percolate were collected.

Preparation of Belladonna Root Percolates with Isopropyl Alcohol

The percolates were prepared in the same manner as above except that 95 per cent isopropyl alcohol replaced ethyl alcohol in the menstruum.

Assay of Belladonna Root Percolates

These percolates were assayed by the official method of the National Formulary, VIII, for Belladonna Root Fluidextract. This procedure is as follows:

"Evaporate 10 cc. of Belladonna Root Fluidextract, accurately measured, on a water bath until the alcohol is all removed. Transfer this extract to 20 cc. of chloroform in a separator, using about 15 cc. of water and 1 cc. of ammonia T.S. to complete the transfer. Shake the mixture vigorously for 1 minute, separate the chloroform layer, and complete the extraction of the alkaloid with successive portions of chloroform. From the combined chloroform extractions in a separator, extract the alkaloids completely by shaking with successive small portions of dilute sulfuric acid (about 1 in 100). Filter the acid solutions successively through a small filter or pledget of cotton into a separator. Add a slight excess of ammonia water, and completely extract the alkaloids from the aqueous layer by shaking with successive portions of chloroform. Evaporate the combined chloroform extractions to dryness on a water bath, and continue the heating for 15 minutes. Redissolve the residue in a small volume of chloroform, evaporate to dryness on a water bath, and continue heating for 15 minutes. Repeat this treatment for the third time. Dissolve the resulting residue in chloroform, add 15 cc. of 0.02 N sulfuric acid, remove the chloroform by evaporation, and titrate the excess acid with 0.02 N sodium hydroxide, using methyl red T.S. as the indicator. Each cc. of 0.02 N sulfuric acid is equivalent to 0.005787 Gm. of the alkaloids of belladonna calculated as atropine."

Assay of Belladonna Root, N. F. VIII

"Moisten 10 Gm. of Belladonna Root, in moderately coarse powder, with a mixture of 3 cc. of stronger ammonia T.S., 20 cc. of ether, and 10 cc. of chloroform, in a small percolator, the outlet of which has been packed with a pledget of purified cotton. Macerate the mixture overnight, and extract by percolating slowly with a mixture of 3 volumes of ether and 1 volume of chloroform. Continue the percolation until the last 3 or 4 cc. of percolate, when evaporated to dryness and the residue dissolved in approximately 0.5 N sulfuric acid and treated with mercuric potassium iodide T.S., shows only a faint turbidity.

"If the volume of liquid obtained is large, reduce it to a convenient volume by evaporating on a water bath.

"Transfer the liquid to a separator, rise the container with one or more small volumes of the solvent, and add the rinsings to the separator. Completely remove the alkaloids from the immiscible solvents by extracting with successive portions of approximately 0.5 N sulfuric acid, filtering each portion drawn off. Render the combined acid solutions distinctly alkaline with ammonia T.S., and completely remove the alkaloids at once by extracting with successive portions of chloroform. Evaporate the combined chloroform extractions to dryness on a water bath, and continue heating for 15 minutes. Redissolve the residue in a small volume of chloroform, evaporate to dryness on a water bath, and continue heating for 15 minutes. Repeat this treatment for the third time. Dissolve the resulting residue in a few cc. of chloroform, add 15 cc. of 0.02 N sulfuric acid, remove the chloroform by evaporation, cool, and titrate the excess acid, with 0.02 N sodium hydroxide, using methyl red T.S. as the indicator. Each cc. of 0.02 N sulfuric acid is equivalent to 0.005787 Gm. of the alkaloids of Belladonna Root, calculated as atropine."

The crude drug was found to contain 0.444 per cent of the alkaloids.

Determination of Total Extractive

The total extractive of the percolates was determined in the following manner:

Fifty cc. of the percolate was placed in a tared beaker, evaporated to dryness on a water bath, and dried to constant weight in an oven at a temperature of 110°C.

The term "dried to constant weight" means that two consecutive weighings do not differ by more than 0.5 mg. per 1 cc. of percolate taken for the determination, the second weighing following an additional hour of drying.

The figures given in Table 1 show that isopropyl alcohol is inferior to ethyl alcohol in the extraction of the alkaloids of Belladonna Root. Isopropyl alcohol is advantageous, however, in that it extracts approximately forty per cent less total extractive. The rather consistent increase in alkaloid content of the successive fractions of the percolate in both cases indicatesthat the drug is not easily extracted with either menstruum.

Table 1. Alkaloid Content and Total Extractive of

Belladonna Root Percolates

Ethyl Alcohol Percolates

Fraction	Alkaloids Extracted Gm. in 200 cc.	Alkaloids Extracted Per Cent	Total Extractive Gm. in 200 cc.
1	0.5600	12.61	11.0348
2	0.6720	15.14	11.9656
3	0.5280	11.89	12.0744
4	0.7460	16.80	11.8016
5	1.0240	23.06	11.5296
Total	3.5300	79.50	58.4060

Isopropyl Alcohol Percolates

Fraction	Alkaloids Extracted Gm. in 200 cc.	Alkaloids Extracted Per Cent	Total Extractive Gm. in 200 cc.
1	0.4880	10.99	6.2588
2	0.5320	11.98	6.7120
3	0.6660	15.00	7.2660
4	0.7240	16.31	7.3848
5	0.7220	16.26	7.1608
Total	3.1320	70.54	34.7824

2. Cinchona

Preparation of Cinchona Percolates with Ethyl Alcohol

To one hundred grams of the ground drug was added a mixture of 130 cc. of ethyl alcohol, 15 cc. of diluted hydrochloric acid, and 50 cc. of water. This was packed in a cylindrical percolator, and macerated for 2 hours. At the expiration of this maceration period, percolation was conducted at a rate of 3 cc. per minute and five 200 cc. fractions of percolate were collected. The extraction was completed with a menstruum consisting of a mixture of two volumes of ethyl alcohol and one volume of water.

Preparation of Cinchona Percolates with Isopropyl Alcohol

The percolates were prepared in the same manner as above except that 95 per cent isopropyl alcohol replaced ethyl alcohol in the menstruum.

Assay of Cinchona Percolates

These percolates were assayed by the official method of the National Formulary, VIII, for Compound Cinchona Tincture which follows:

"Accurately measure 50 cc. of Compound Cinchona Tincture and evaporate it, at a temperature not exceeding 100°, to a volume of about 10 cc. Add sufficient asbestos fiber or paper pulp to absorb the liquid, and continue the evaporation to dryness. Transfer the residue to a flask or bottle, add 200 cc., accurately measured at room temperature, of ether-chloroform mixture (ether 4 volumes, chloroform 1 volume) and sufficient ammonia T.S. (which may be used to rinse out the adhering portions of the Tincture from the evaporating dish) to render the mixture strongly alkaline. Securely stopper the container and shake mechanically during 1 hour, or intermittently during 2 hours, and then allow the mixture to stand overnight. Again shake the mixture intermittently for 30 minutes, allow to settle, quickly decant 160 cc. (representing 40 cc. of the Tincture) of the approximately clear liquid. Filter this into a separator and wash the measuring vessel with sufficient of the ether-chloroform mixture, adding the rinsings to the filter. Extract the alkaloids from the clear liquid with acidified water, using sufficient diluted sulfuric acid to render the contents of the separator and each extract distinctly acid to litmus paper. Pass the acid extracts in succession through a moistened, double filter into a second separator. Render the combined liquids distinctly alkaline with stronger ammonia T.S., and extract with chloroform. Pass the chloroform extracts through a double filter, which is kept saturated with chloroform, into a suitable, tared receptacle. Evaporate the chloroform on a water bath, dry the residue to constant weight at 100° and weigh. The weight multiplied by 2.5 indicates the weight of alkaloids in 100 cc. of the Compound Cinchona Tincture."

Assay of Cinchona, N. F. VIII

"Place 5 Gm. of Cinchona, in fine powder, and 15 cc. of 3 per cent hydrochloric acid in a 500 cc. flask and heat the mixture on a water bath for 1 hour. Cool and add 200 cc. of ether-chloroform solution (ether 3 volumes, chloroform 1 volume) and 10 cc. of stronger ammonia T.S. Stopper the flask tightly and shake it for 1 hour in a mechanical shaker. Allow the mixture to stand overnight, again shake it for 30 minutes, and then allow the drug to settle. (If the supernatant liquid is not clear, add a few cc. of distilled water, again shake the contents of the flask vigorously, and allow the drug to settle.)

"Quickly decant 160 cc. of the clear, ether-chloroform solution, measured at approximately the same temperature as the original ether-chloroform solution and representing 4 Gm. of the drug. Transfer the solution to a separator, rinse the measuring vessel with a small quantity of the original menstruum, and add the rinsings to the separator. Completely extract the alkaloids with approximately 5 per cent sulfuric acid, and collect the acid solution of the alkaloids in a second separator.

"Make the acid solution strongly alkaline with ammonia T.S., and completely extract the alkaloids with chloroform. Evaporate or distill the chloroform in a tared beaker or flask and dry the alkaloidal residue to constant weight at 100°. The weight obtained, multiplied by 25, represents the per cent of the alkaloids of Cinchona in the drug."

Determination of Total Extractive

The total extractive of the percolates was determined in the same manner as for Belladonna Root Percolates, page 8.

The figures in Table 2 show that isopropyl alcohol is superior to ethyl alcohol in the extraction of the alkaloids of cinchona. However, isopropyl alcohol extracted approximately one per cent more of the total extractive. This slight increase in the amount of total extractive is apparently due to the increased content of alkaloids. Also, the consistent decrease in alkaloid content and total extractive of the successive fractions of percolate in both cases indicate that the drug is easily extracted with either menstruum.

Table 2. Alkaloid Content and Total Extractive of Cinchona Percolates

Ethyl Alcohol Percolates

Fraction	Alkaloids Extracted Gm. in 200 cc.	Alkaloids Extracted Per Cent	Total Extractive Gm. in 200 cc.
1	3.6550	31.81	22.1184
2	2.1440	18.66	9.6488
3	1.6325	14.21	5.2616
4	0.7005	6.10	2.1560
5	0.5065	4.41	1.3136
Total	8.6385	75.19	40.4984

Isopropyl Alcohol Percolates

Fraction	Alkaloids Extracted Gm. in 200 cc.	Alkaloids Extracted Per Cent	Total Extractive Gm. in 200 cc.
1	3.5320	30.74	16.8876
2	2.7460	23.90	11.3080
3	1.7270	15.03	7.4616
4	1.2785	11.13	3.4584
5	0.7585	6.60	1.9908
Total	10.0420	87.40	41.1064

3. Nux Vomica

Preparation of Nux Vomica Percolates with Ethyl Alcohol

One thousand grams of the ground drug was mixed with a sufficient quantity of a menstruum consisting of 100 cc. of acetic acid, 150 cc. of water, and 750 cc. of ethyl alcohol, to render it evenly and distinctly damp. The dampened drug was allowed to stand for 15 minutes after which it was packed firmly in a cylindrical percolator and the remainder of the menstruum was added. When the liquid began to drop from the percolator, the lower orifice was closed, the percolator covered, and the drug allowed to macerate for 48 hours. At the expiration of this maceration period, percolation was conducted at a rate of 1 cc. per minute and five 200 cc. fractions of percolate were collected. The extraction was completed with a menstruum consisting of three volumes of ethyl alcohol and one volume of water.

Preparation of Nux Vomica Percolates with Isopropyl Alcohol

The percolates were prepared in the same manner as above except that 95 per cent isopropyl alcohol replaced ethyl alcohol in the menstruum.

Assay of Nux Vomica Percolates

These percolates were assayed by the official method of the National Formulary, VIII, for Nux Vomica Fluidextract as follows:

"Transfer 10 cc. of Nux Vomica Fluidextract, accurately measured, to a separator containing about 30 cc. of chloroform, add 5 cc. of water and 5 cc. of ammonia T.S., and shake well for 1 minute. Draw off the separated chloroform solution, and complete the extraction of the alkaloids from the alkaline liquid by shaking with successive portions of chloroform. Combine the chloroform solutions. Add about 40 cc. of approximately 1 N sulfuric acid to the separator, and shake the mixture gently for 5 minutes, then allow the liquids to separate, and draw off the acid into another separator. Repeat the extraction with successive portions of the acid, until the ether solution is completely extracted.

"To the combined acid solutions in the separator add a small piece of red litmus paper and 50 cc. of chloroform, and follow with sufficient ammonia T.S. to render the aqueous layer alkaline, and after gently shaking, add 2 or 3 cc. more of the ammonia T.S. Now shake the mixture thoroughly, but gently, for about 10 minutes, and allow the liquids to separate. Draw off the chloroform into a container, and repeat the extraction with additional portions of chloroform until all of the alkaloid is extracted. Extract the combined chloroform solutions with successive portions of approximately 1 N sulfuric acid until completely extracted. Then render the combined acid solutions alkaline with ammonia T.S., add 2 or 3 cc. more of the ammonia T.S., and completely extract the alkaloids with successive portions of chloroform.

"Carefully evaporate the combined chloroform extracts to dryness on a water bath, dissolve the residue by warming with 15 cc. of approximately 3 per cent sulfuric acid, cool to 25°, and add 3 cc. of a mixture of equal parts of nitric acid and a 5 per cent solution of sodium nitrite in distilled water. Thoroughly stir this mixture, and allow it to stand for exactly 10 minutes at room temperature. At the expiration of this period, pour the red solution at once into a separator containing 50 cc. of chloroform and 15 cc. of sodium hydroxide solution (1 in 10), and rinse the flask with distilled water, adding the rinsings to the separator. Add sufficient sodium hydroxide solution (1 in 10) to the contents of the separator to render it distinctly alkaline to litmus paper, and then add a few cc. more of the sodium hydroxide solution. Shake the mixture gently for 10 minutes and allow the liquids to separate. Draw off the chloroform layer into another separator and repeat the extraction with additional portions of chloroform until the alkaloid is completely removed. Add 10 cc. of distilled water to the combined chloroform extract, shake the mixture gently, and add a small piece of red litmus paper. The litmus paper should indicate not more than a slight alkalinity. If the water, after shaking with the chloroform, is strongly alkaline, draw off the chloroform into another separator, and shake it with another 10 cc. of distilled water. Draw off the chloroform, passing it through a filter paper moistened with chloroform, into a container. Wash the filter paper with warm chloroform, and add these rinsings to the container. Now shake the combined aqueous extract with 5 cc. of chloroform and draw off this chloroform, passing it through the chloroform-moistened filter paper into the main chloroform solution.

"Evaporate the combined chloroform extracts very carefully on a water bath nearly, but not quite, to dryness. Add to the moist residue 7 cc. of 0.1 N sulfuric acid, accurately measured, follow with 30 cc. of distilled water, and heat the mixture on a water bath until the alkaloid is dissolved and the odor of chloroform is dissipated. Cool to room temperature and titrate the excess acid with 0.02 N sodium hydroxide, using 1 drop of methyl red T.S. as the indicator. Each cc. of 0.1 N sulfuric acid is equivalent to 0.03344 Gm. of $C_{21}H_{22}O_2N_2$."

Assay of Nux Vomica, N. F. VIII

"Place 15 Gm. of Nux Vomica, in coarse powder, in a flask or bottle, add 150 cc., measured at 25°C of a mixture of 3 volumes of ether and 1 volume of chloroform, tightly stopper the flask, shake the mixture, and allow it to stand for about 2 minutes. Then add 10 cc. of stronger ammonia T.S., stopper the flask tightly, shake frequently, but gently, during 1 hour, and allow the mixture to stand overnight at a temperature not over 25°. Then quickly transfer to a separator exactly 100 cc. of the liquid, representing 10 Gm. of Nux Vomica, rinse the measuring vessel with a little chloroform, and add the rinsings to the separator. Add about 40 cc. of approximately 1 N sulfuric acid to the separator, and shake the mixture gently for 5 minutes, then allow the liquids to separate, and draw off the acid into another separator. Repeat the extraction with successive portions of the acid, until the ether solution is completely extracted.

"To the combined acid solutions in the separator add a small piece of red litmus paper and 50 cc. of chloroform, and follow with sufficient ammonia T.S. to render the aqueous layer alkaline, and after gently shaking, add 2 or 3 cc. more of the ammonia T.S. Now shake the mixture thoroughly but gently, for about 10 minutes, and allow the liquids to separate. Draw off the chloroform into a container, and repeat the extraction with additional portions of chloroform until all of the alkaloid is extracted. Extract the combined chloroform solutions with successive portions of approximately 1 N sulfuric acid until completely extracted. Then render the combined acid solutions alkaline with ammonia T.S., add 2 or 3 cc. more of the ammonia T.S., and completely extract the alkaloids with successive portions of chloroform.

"Carefully evaporate the combined chloroform extracts to dryness on a water bath, dissolve the residue by warming with 15 cc. of approximately 3 per cent sulfuric acid, cool to 25°, and add 3 cc. of a mixture of equal parts of nitric acid and a 5 per cent solution of sodium nitrite in distilled water. Thoroughly stir this mixture, and allow it to stand for exactly 10 minutes at room temperature. At the expiration of this period, pour the red solution at once into a separator containing 50 cc. of chloroform and 15 cc. of sodium hydroxide solution (1 in 10), and rinse the flask with distilled water, adding the rinsings to the separator. Add sufficient sodium hydroxide solution (1 in 10), to the contents of the separator to render it distinctly alkaline to litmus paper, and then add a few cc. more of the sodium hydroxide solution. Shake the mixture gently for 10 minutes and allow the liquids to separate. Draw off the chloroform layer into another separator and repeat the extraction with additional portions of chloroform until the alkaloid is completely removed. Add 10 cc. of distilled water to the combined chloroform extract, shake the mixture gently, and add a small piece of red litmus paper. The litmus paper should indicate not more than a slight alkalinity. If the water, after shaking with the chloroform, is strongly alkaline, draw off the chloroform into another separator, and shake it with another 10 cc. of distilled water. Draw off the chloroform, passing it through a filter paper moistened with chloroform, into a container. Wash the filter paper with warm chloroform, and add these rinsings to the container. Now shake the combined aqueous extract with 5 cc. of chloroform and draw off this chloroform, passing it through the chloroform-moistened filter paper into the main chloroform solution.

"Evaporate the combined chloroform extracts very carefully on a water bath nearly, but not quite, to dryness. Add to the moist residue 7 cc. of 0.1 N sulfuric acid, accurately measured, and follow with 30 cc. of distilled water, and heat the mixture on a water bath until the alkaloid is dissolved and the odor of chloroform is dissipated. Cool to room temperature, and titrate the excess acid with 0.02 N sodium hydroxide, using 1 drop of methyl red T.S. as the indicator. Each cc. of 0.1 N sulfuric acid is equivalent to 0.03344 Gm. of $C_{21}H_{22}O_2N_2$."

The crude drug was found to contain 1.12 per cent of strychnine.

Determination of Total Extractive

The total extractive of the percolates was determined in the same manner as for Belladonna Root Percolates, page 8.

Fat Determination of Nux Vomica Percolates

The fat content of the percolates was determined in the following manner:

The preparations were chilled to a temperature of 10°C. for a period of 7 days. The separated fat was then removed by filtration while cold, all apparatus being chilled to the same temperature before filtration was conducted. A 50 cc. sample of the filtrate was then evaporated to dryness on a water bath, and the residue dried to constant weight in an oven at a temperature of 110°C. The quantity of fat in a 50 cc. sample of the percolate was then obtained by subtracting the amount of extractive obtained here from the quantity of total extractive previously determined.

The results in Table 3 show that ethyl alcohol is a better menstruum than isopropyl alcohol in the extraction of strychnine from Nux Vomica, and also extracts a smaller quantity of fat. However, the isopropyl alcohol percolates yielded approximately sixteen per cent less total extractive. Since the National Formulary requires the removal of fat by chilling prior to completion of official preparations, the large amount of inert fat extracted by isopropyl alcohol can be easily removed and actually would result in a lower total extractive yield. In both cases there was an increase in the strychnine content of the successive fractions of percolate indicating that this alkaloid is extracted with difficulty from Nux Vomica with either menstruum.

Table 3. Strychnine Content, Total Extractive, and Fat Content of Nux Vomica Percolates

Ethyl Alcohol Percolates

Fraction	Strychnine Extracted Gm. in 200 cc.	Strychnine Extracted Per Cent	Total Extractive Gm. in 200 cc.	Fat Gm. in 200 cc.
1	1.5460	13.80	12.6576	1.0216
2	1.7160	15.32	14.3632	1.7752
3	1.8540	16.55	15.8732	1.9332
4	2.1400	19.11	18.1788	2.2508
5	2.3220	20.73	18.0260	1.4940
Total	9.5780	85.51	79.0988	8.4748

Isopropyl Alcohol Percolates

Fraction	Strychnine Extracted Gm. in 200 cc.	Strychnine Extracted Per Cent	Total Extractive Gm. in 200 cc.	Fat Gm. in 200 cc.
1	1.4520	12.96	11.2184	1.3784
2	1.4160	12.64	11.6268	1.8988
3	1.4640	13.07	13.2600	2.6920
4	1.6320	14.57	15.8140	3.8840
5	1.9640	17.54	14.4604	1.8644
Total	7.9280	70.78	66.3796	11.7176

4. Ginger

Preparation of Ginger Percolates with Ethyl Alcohol

One thousand grams of the ground drug was mixed with a sufficient quantity of a mixture of nine volumes of ethyl alcohol and one volume of water to render it evenly and distinctly damp. This required 700 cc. of solution. The dampened drug was allowed to stand for 15 minutes. It was then packed firmly in a cylindrical percolator and a sufficient quantity of menstruum (nine volumes of ethyl alcohol and one volume of water) was added to saturate the drug and leave a stratum above. When the liquid began to drop from the percolator, the lower orifice was closed, the percolator covered, and the drug allowed to macerate over-night. At the expiration of this maceration period, percolation was conducted at a rate of 1 cc. per minute and five 200 cc. fractions of percolate were collected.

Preparation of Ginger Percolates with Isopropyl Alcohol

The percolates were prepared in the same manner as above except that 95 per cent isopropyl alcohol replaced ethyl alcohol in the menstruum.

Assay of Ginger Percolates

These percolates were assayed by the official method of the United States Pharmacopoeia, XIII, for Ginger Fluidextract. This procedure is as follows:

"Place 20 cc. of Ginger Fluidextract in a 200 cc. beaker, and evaporate on a water bath until there is no longer any odor of alcohol. Remove the beaker from the bath, and add 50 cc. of ether. Stir the contents of the beaker with a stirring rod to dissolve the soluble resin, and decant the ether through a dry, 9 cm. filter into a tared, 200 cc. beaker. Repeat the extraction two or three times, using 50 cc. portions of ether. Wash the filter with a small amount of ether, and evaporate the combined ether extracts on a water bath until the odor of ether is no longer perceptible. Finally dry in a desiccator over sulfuric acid for 18 hours, and then weigh."

Assay of Ginger, U. S. P. XIII

"Place 20 Gm. of Ginger, in fine powder, and accurately weighed, in an extraction thimble of a Soxhlet or similar extractor, and extract with ether for 6 hours. Evaporate the ether extract on a steam bath until the odor of ether is no longer perceptible, then dry the residue in a desiccator over sulfuric acid for 18 hours."

The crude drug was found to contain 3.95 per cent of ether-soluble extractive.

Determination of Total Extractive

The total extractive of the percolates was determined in the following manner:

Fifty cc. of the percolate was placed in a tared beaker, evaporated to dryness on a water bath, and the residue dried in a desiccator over sulfuric acid for 18 hours.

Determination of Volatile Extractive

Because the official assay for Ginger Fluidextract is a measure of the ether-soluble constituents consisting of volatile oil and non-volatile resinous material, a determination of the volatile extractive was made also. This was done to show the amount of volatile and non-volatile material in the ether-soluble extractive.

The volatile extractive of the percolates was determined in the following manner:

Fifty cc. of the percolate was placed in a tared beaker, evaporated to dryness on a water bath, and dried to constant weight in an oven at a temperature of 110°C.

The difference between the weight of the total extractive previously determined and the non-volatile residue obtained here represents the volatile extractive content.

The results in Table 4 show that ethyl alcohol is superior to isopropyl alcohol in the extraction of the ether-soluble constituents as well as the volatile constituents of Ginger. The consistent decrease of these active constituents from fraction to fraction of ethyl alcohol percolates indicates that they are more easily removed by ethyl alcohol than by isopropyl alcohol. However, the ethyl alcohol percolates yielded approximately thirty-five per cent more total extractive as compared to the isopropyl alcohol percolates.

Table 4. Ether-Soluble Extractive, Total Extractive, and Volatile Extractive of Ginger Percolates

Ethyl Alcohol Percolates

Fraction	Ether-Soluble Extractive Gm. in 200 cc.	Ether-Soluble Extractive Per Cent	Volatile Extractive Gm. in 200 cc.	Total Extractive Gm. in 200 cc.
1	12.9360	32.75	4.5200	14.2076
2	8.4970	21.51	4.2280	11.3044
3	7.0750	17.91	2.7924	9.3580
4	5.7470	14.55	3.2576	8.6016
5	4.1600	10.53	2.3332	6.7864
Total	38.4150	97.25	17.1312	50.2580

Isopropyl Alcohol Percolates

Fraction	Ether-Soluble Extractive Gm. in 200 cc.	Ether-Soluble Extractive Per Cent	Volatile Extractive Gm. in 200 cc.	Total Extractive Gm. in 200 cc.
1	6.3190	16.00	2.7124	7.5736
2	7.4940	18.97	2.8588	8.7324
3	6.8280	17.29	2.2784	7.5512
4	2.8810	7.29	1.3680	4.1216
5	3.4970	8.85	1.8836	4.7424
Total	27.0190	68.40	11.1012	32.7212

CONCLUSIONS

1. A quantitative study has been made employing both ethyl and isopropyl alcohols as menstrua for the extraction of Belladonna Root, Cinchona, Nux Vomica and Ginger.

2. Ethyl alcohol is superior for the extraction of the active principles of Belladonna Root. However, isopropyl alcohol removed approximately forty per cent less total extractive.

3. Isopropyl alcohol proved to be better than ethyl alcohol for the extraction of Cinchona alkaloids. Both menstrua yielded about the same quantity of total extractive.

4. In the case of Nux Vomica, ethyl alcohol extracted a larger quantity of strychnine, more total extractive, and less fat than did isopropyl alcohol.

5. Ethyl alcohol excelled isopropyl alcohol in the extraction of Ginger. The former removed more ether-soluble extractive, more volatile extractive, and more total extractive than did isopropyl alcohol.

6. The physical appearance of the preparations made with isopropyl alcohol were identical to those made with ethyl alcohol.

7. The odor and taste of isopropyl alcohol are minor objections when compared with ethyl alcohol.

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