

phen and phenobarbital in mixtures and by a combination of these methods with A. O. A. C. procedures, determinations of as many as four of the constituents in the same mixture have been made. The recoveries by the recommended methods range from about 98 per cent to about 104 per cent.

Precipitation of the aminopyrine as the double salt with cadmium iodide or with mercuric chloride was not entirely satisfactory. Attempts to separate the several substances by the differences in their solubilities in various solvents were not satisfactory.

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The Determination of Camphor in Camphor Liniment

An Accurate and Simplified Volatilization Method

By Solomon M. Berman*

The last three revisions of the U. S. Pharmacopœia have presented as many different assay methods for Liniment of Camphor. The U. S. P. IX method involved removal of the camphor from the oily vehicle by means of hot alcohol vapor, the camphor in the distillate being measured polariscopically. The U. S. P. X assay was essentially that proposed by Miller (1) who stated that 90 minutes heating at 110° in a platinum dish was sufficient to volatilize the camphor, without decomposing the oil. He criticized the earlier method as impractical. Previously, Cook (2) had suggested a 3-hour heating at 100° for removing 97.5% of the camphor in a 20% liniment, with a suitable correction factor. Lothian (3) recommended the use of petri dishes and heating on the water-bath for one hour. He noted that the residue gained in weight on further heating, while the oil, used in preparing the liniment, did not gain. He concluded that blanks were not dependable.

In a series of papers, Poe and others (4) made a detailed report of studies on the assay of Camphor Liniment. They found that the U. S. P. X method gave low results on the official liniment as well as on liniments prepared from the other common fixed vegetable oils. They also observed that aluminum dishes gave the lowest results in any series of determinations, while glass dishes were relatively satisfactory and tin or lead vessels were best. These workers found that the most serious error in the volatilization procedure was due to oxidation of the oil at the temperature used, the effect increasing with time and more pronounced when aluminum vessels were employed. Subsequent studies of heating *in vacuo*, in nitrogen, in carbon dioxide, and in the presence of anti-oxidants, led to their

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recommending the use of the vacuum oven at 115–120° for two hours, as yielding results nearest to theoretical. Their data largely support this recommendation, but also show decomposition of oil under those conditions amounting to 0.12%, which corresponds to 0.48% of the camphor in a 20% liniment.

The method at present official in the U. S. P. XI is also official (with slightly different wording) in the N. F. VI for the assay of Ampuls of Camphor, and is due to Grantham and his collaborators (5). Their results are in essential accord with those of Poe (4) for the parallel method, and for eleven determinations ranged from 98.2% to 102.5% recovery, with an average of 99.9%. It should be noted that the collaborating analysts reported from three to six hours as the necessary heating period.

Overbye and Schoetzow (6), using various 20% liniments, found by this method amounts of camphor varying from 18.63% to 19.89%, after correcting for loss in drying the oils alone. The range of results corresponds to recovery of from 93.15% to 99.45% of the camphor content. They concluded that a blank was needed on the particular oil used in preparing the liniment.

A method was sought which would be rapid and accurate, and which would not cause any significant change in the oily vehicle of the Camphor Liniment. The present report is believed to describe such a method.

EXPERIMENTAL

Three batches of Camphor Liniment were prepared, corresponding to the upper and lower limits as well as to the nominal 20% formula requirement of the U. S. P. XI (range—19% to 21% camphor). The camphor used was evidently a synthetic product in view of its small optical activity. (Five grams in 50 cc. alcohol showed 3.8° in 100-mm. tube.) The cottonseed oil was a commercial edible oil, meeting the U. S. P. requirements, and about two year old.

The U. S. P. method of preparation was followed in each case, the weighed camphor (by difference) being added to the warm cottonseed oil, contained in an Erlenmeyer flask; the flask tightly stoppered with a foil-lined cork and agitated on a rotating machine for several hours until a perfectly clear solution was obtained. Liniment *A* consisted of 81.0072 Gm. of oil and 19.5761 Gm. of camphor, correspond-

ing to 19.463% of camphor. Liniment *B* consisted of 79.9993 Gm. of oil and 20.1240 Gm. of camphor or 20.099% of camphor. Liniment *C* consisted of 79.0031 Gm. of oil and 20.9234 Gm. of camphor or 20.939% of camphor.

In the initial experiment, only Liniments *A* and *B* were used. Approximately 5-cc. samples were weighed into petri dishes of 90 mm. diameter, each dish having as tare a similar petri dish, carried along with it through the conditions of the analysis. A 4-hole water-bath was used, so only two liniments and their tares could be run simultaneously. The bath was electrically heated, the water temperature being 99–100°. A draft of the hood window-sash type was not available, a medium size electric fan operating at moderate speed in fixed direction being used instead. The air current was mainly across the dish, but partly into it. A 30-minute heating was followed by 15-minute heatings to constant weight, the weighings being made after each heating. Five to 10 minutes standing in a current of air, preferably on a conducting surface, was sufficient to bring the vessels and contents to room temperature. The results are given in Table I.

Table I.—Recovery of Camphor Based on Loss in Weight of Liniment

Lini- ment	Sample Wt., Gm.	In Petri Dishes On Water- Bath with Air Draft		
		Recovered, %		
		30 Min.	45 Min.	60 Min.
<i>A</i>	4.5445	99.49	99.85	99.85
<i>A</i>	4.8950	99.42	99.88	99.84
<i>B</i>	4.9458	98.72	99.80	99.86
<i>B</i>	4.5009	99.34	99.83	99.81

In another series, an 8 hole gas-fired water-bath was used, duplicate samples of the 3 liniments being run concurrently, with a single petri dish as tare. A considerable differential existed in the degree of draft afforded to the dishes at opposite ends of the bath. The size of sample was permitted to vary from 3.0 to 5.3 Gm. In no case did the oil completely cover the floor of the dish even when warm. The water-bath temperature was 99°. A maximum thermometer, or other device for measuring the highest temperature attained by the oil samples, was not available. However, a beaker of oil immersed in the steam with frequent agitation showed a maximum of 95° in the 30-minute period. It is estimated that the temperature of the liniment samples varied between 70° and 90° depending upon their nearness to the fan. The data are given in Table II.

Table II.—Recovery of Camphor Based on Loss in Weight of Liniment

Lini- ment	Sample Wt., Gm.	In Petri Dishes on Water- Bath with Air Draft		
		Recovered, %		
		30 Min.	45 Min.	60 Min.
<i>A</i>	4.6919	97.82	99.42	99.62
<i>B</i>	4.5423	98.59	99.63	99.57
<i>C</i>	5.3062	96.44	99.41	99.68
<i>A</i>	4.0464	98.17	99.24	99.38
<i>B</i>	3.2661	99.55	99.64	99.60
<i>C</i>	3.0040	99.27	99.54	99.56

Aluminum dishes similar in size to the petri dishes, and of rather heavy construction, were tried. They were found objectionable because they were too hot to be handled safely with the fingers on removal from the bath, the use of tongs being inadvisable as they might readily come in contact with the oil. The experiment was abandoned after 45 minutes of heating, 9 determinations showing from 97.67% to 100.22%, before constant weight was attained.

Test was then made of light weight aluminum dishes having a diameter of 57 mm. (against 90 mm. for the petri dishes), a depth of 10 mm., and weighing about 4 Gm. In contrast to the larger aluminum dishes, these were found very convenient. The size of the sample was reduced to about 3 Gm. so as to barely cover the floor of the vessel. In the first two trials reported in Table III, the tare was carried through the conditions of the assay, while in the rest the tare remained on the balance pan. The bath temperature was 100°.

Table III.—Recovery of Camphor Based on Loss in Weight of Liniment

Lini- ment	Sample Wt., Gm.	In Aluminum Dishes on Water- Bath with Air Draft		
		Recovered, %		
		30 Min.	45 Min.	60 Min.
B	3.0215	99.87	99.86
C	2.8546	99.98	99.91
A	2.6831	98.98	99.85	99.83
A	2.9176	100.03	100.18	100.13
B	2.6549	99.59	99.94	99.93
B	2.8975	96.61	99.20	99.75
C	3.2998	97.71	99.62	100.02
C	3.1048	99.49	99.77	99.74

A partial study was made of the effect a lower bath temperature would have. An effort was made to equalize the fan effects for this trial.

Table IV.—Recovery of Camphor Based on Loss in Weight of Liniment

Lini- ment	Sample Wt., Gm.	In Petri Dishes on 95° Water- Bath with Air Draft			
		Recovered %			
		30 Min.	45 Min.	60 Min.	75 Min.
A	5.3092	87.4	94.6	99.13	99.74
A	5.3885	81.6	93.5	98.51	99.69
B	5.2293	95.5	98.9	99.48	99.68
B	4.9084	94.0	97.6	99.18	99.55
C	4.5455	97.0	99.1	99.66	99.76
C	4.6948	97.9	99.5	99.69	99.70

Beakers of 150-cc. capacity and 60-mm. diameter were also tried, immersed in the 95° bath. A 30-minute heating gave 88% to 94% recovery on four samples. Their use indicated no advantage, and was not tested further.

Table V.—Comparative Assays of Commercial Camphor Liniment

Lini- ment	In Petri Dishes on Water- Bath with Air Draft			U. S. P. XI, 2 Hours 110°—CO ₂	
	Recovered, %				
	30 Min.	45 Min.	60 Min.		
(1)	17.14	17.20	17.19	17.14,	17.13 ^a
(2)	18.78	18.78	...	18.77,	18.79 ^a
(3)	25.13	25.09	...	25.21,	25.22 ^b
(3)	24.00	25.02	25.07
(4)	17.47	17.65	17.60	17.59,	17.58 ^b
(5)	16.62	16.84	16.88	16.93,	16.93 ^b

^a Routine analyses by D. W. McLaren.

^b Routine analyses by R. L. Herd.

The new method was then used to assay five commercial liniments, in comparison with the U. S. P. XI assay conducted at the same time.

In the foregoing experiments necessity dictated the use of an electric fan. When a steam-bath with cross draft became available, a comparison was made of the two types of ventilation involved. The cross draft was produced by a blower creating suction through a slot at the bottom of the baffle forming the back of the hood which contained the steam-bath. The front of the hood was completely open, the resulting draft being more moderate than if a sash had been used, and quite lacking the blowing effect of the fan. Table VI shows a single comparison, using a commercial liniment and petri dishes.

Table VI.—Comparative Assay of Commercial Camphor Liniment

	In Petri Dishes on Steam-Bath Recovered, %	
	30 Min.	45 Min.
Hood draft	19.74	19.75
Electric fan	19.73	19.75

Six samples of cottonseed oil were treated under most rigorous assay conditions, three of the samples receiving collaborative attention. The collaborators were instructed to:

"Weigh accurately from 2 to 5 Gm. of sample in a shallow, flat-bottomed dish of thin-walled construction and such size that the sample barely covers the floor of the dish. Use a similar dish as tare, and carry through the conditions of analysis. Heat the dish on a boiling water- or steam-bath for 30 minutes, in a strong current of air. Cool to room temperature and weigh. Heat as before, for 15 minutes, and again weigh. Repeat this procedure two more times, so that the total heating time is 75 minutes."

Four of the collaborators were directed to use glass dishes, and all chose petri dishes. The two other collaborators used light aluminum dishes (5.7 cm. diameter) to check on any possible difference due to vessels of this material.

Table VII.—Maximum Gain in Weight of Cottonseed Oils

Collab. ^a	In Petri Dishes ^b on Steam-Bath with Air Draft				
	Gain in Wt., %				
	Oil No. 1	Oil No. 2	Oil No. 3	Oil No. 4	Oil No. 5
H. R.	0.03	0.02	0.01
C. F. B.	0.02	0.02	0.02
J. D. C.	0.04	0.04	0.02
D. M. T.	0.03	0.03	0.05
	0.02	0.05	0.04
	0.03	0.01	0.00
	0.03	0.00	0.03
In Aluminum Dishes ^c					
R. L. H.	0.05	0.03	0.06
	0.03	0.05	0.04
S. M. B.	0.02	0.02	0.02	0.03	0.01
	0.02	0.00	0.05	0.01	0.01
Average	0.03	0.02	0.03	0.02	0.01

^a Collaborators: Harry Rogavitz, New York; C. F. Bruening, Baltimore; James D. Curphey, Philadelphia; D. M. Taylor, Philadelphia; R. L. Herd, Buffalo; the author all members U. S. Food and Drug Administration.

^b Sample weight from 2.99 Gm. to 3.71 Gm.

^c Sample weight from 2.88 Gm. to 4.46 Gm.

The first four cottonseed oils of Table VII were commercial oils from as many different manufacturers of Camphor Liniment and had been bought for liniment vehicles. One of them was also intended for preparation of Ampuls Camphor in Oil N. F. (The N. F. VI permits use of "any bland fixed oil.") Oil No. 5 was the base of the laboratory-made liniments. A 5.1-Gm. sample of this oil heated for a single 45-minute period gained 0.03% in a petri dish.

The sixth oil was the residue from the commercial Camphor Liniment of Table VI. The residue from the "hood draft" treatment gained 0.07% after two hours' further heating, while the residue from the "fan" treatment gained 0.17% after two and one-half hours' heating preceded by an equal period of exposure at room temperature. The over-all exposure to air of the collaborative samples was four to five hours.

DISCUSSION

So far as the writer has been able to ascertain, the use of efficient ventilation as a means of volatilizing the camphor at oil temperatures below 100° has not previously been suggested. Many laboratories have ovens equipped with fans in constant motion. It is fairly certain that where such was the case the U. S. P. X method gave good results. The foregoing tables show clearly that a high order of accuracy may be attained in the assay of Liniment of Camphor by simply heating the liniment in a current of air on a bath maintained at close to 100°, and weighing the residue at relatively short intervals. A difference between two successive weighings of less than 1 mg. per Gm. sample would be a sufficient assurance that the volatilization of the camphor was as complete as usually necessary. Under well-standardized conditions a single 30-minute heating should be enough to yield better than 99% recovery. In view of the fact that such conditions would not be universally obtainable it is well that no error, and but little inconvenience, is introduced by heating to constant weight.

The insignificance, analytically, of the effect of the assay conditions on the cottonseed oil base is shown in three ways: (a) the very slight gain in weight of five different cottonseed oils subjected to assay conditions, both in glass and aluminum, (b) the uniformly excellent recovery of camphor from three laboratory made liniments and

(c) the very slight gain in weight of the residues on continued heating. It must be inferred that the oxidative changes noted by previous workers were associated with the higher temperatures and longer heating periods they employed.

Since it appeared more informative to report camphor recovery than to report per cent camphor in any given liniment, it was necessary to recalculate the data of others for comparison. Obviously, in the case of the commercial samples, where the true camphor content was unknown, only the found per cent camphor in the liniment could be shown.

The tabulated comparison of results by the proposed method and by the U. S. P. XI assay shows that the two procedures can yield results in excellent agreement. From a practical aspect, however, the proposed method presents several important improvements: (a) it enables a large number of determinations to be made concurrently; (b) it makes unnecessary the monopoly of a relatively expensive and busy piece of equipment, the electric oven, for two hour periods; (c) it eliminates the need for accessories, such as drying tubes, delivery tubes and carbon dioxide generator; and (d) it furnishes the analyst with an objective record of the completed loss of volatile matter from his sample.

SUMMARY

(1) The literature on volatilization methods for assaying Liniment of Camphor is briefly reviewed.

(2) A simplified method is described, consisting essentially of heating the Liniment of Camphor on a boiling water-bath, in a current of air, to constant weight.

(3) A number of results are tabulated to show the rapidity and dependability of the method under varying conditions. Twenty-four determinations averaged 99.75% recovery.

(4) It is shown that aluminum vessels may be used instead of glass.

(5) The new method is compared with the official assay of the U. S. P. XI (and of the N. F. VI for Ampuls of Camphor) and certain practical advantages are noted.

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Determination of Iodine in Thyroid

Comparison of a Rapid Method with the U. S. P. XI Method

By Charles E. Nicklaus* and Nelson Tippett*

Previous investigations have emphasized the need of an assay method for Thyroid, that is both quick and accurate. Tippett (1) simplified the U. S. P. XI Method and increased the accuracy by consideration of a titration time factor. The one inherent disadvantage of the alkaline ash method remained however—one assay taking the better part of a working day. Nicklaus has adapted the Matthews, Curtis and Brode modification of the Leipert procedure (2) to Thyroid products and obtained a method having the triple advantage of speed, accuracy and manipulative ease. In substance this method depends upon an initial chromium trioxide oxidation and distillation in a special, but quite simple apparatus, and a subsequent titration with hundredth-normal sodium thiosulfate, the complete procedure requiring a maximum time of one hour. Over a period of six months, numerous assays by each method have shown practically no variation. For the appended table a representative few have been chosen.

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If care is taken to use reagent grade chemicals, no blank is needed.

EXPERIMENTAL

Preparation of Reagents.—30*N* Chromium Trioxide: Dissolve 165.0 Gm. chromium trioxide in 100 cc. water.

N Potassium Carbonate: Dissolve 14.0 Gm. of the salt in 100 cc. water.

0.2*M* Potassium Permanganate: Dissolve 3.16 Gm. in 100 cc. water.

1.5*N* Sodium Nitrite: Dissolve 10.4 Gm. in 100 cc. water.

5*M* Urea Solution: Dissolve 30.0 Gm. in 100 cc. water.

Determination of Iodine.—Weigh accurately about 0.5 Gm. of Thyroid and place it in a 500-cc. Kjeldahl flask (containing an antibump) and wash down the inside of the flask with about 10 cc. of distilled water. If the assay is to be run on a tablet, a representative number are weighed and powdered and an aliquot part of the powder, equivalent to about 0.5 Gm. of Thyroid, is treated in the same manner. No trouble is experienced with tablets of any grainage.

With constant rotation of the flask, slowly add 10 cc. of 30*N* chromium trioxide, and after the reaction has subsided add slowly 50 cc. of sulfuric acid. Place the flask in a ring stand and fit it with a three-hole rubber stopper containing a 0–220°C. thermometer and two tubes for ventilation. Heat the contents at a temperature of 210–220°C. until the liquid acquires a clear bluish green coloration; when the bubbles cease to aggregate on the surface of the liquid, oxidation is complete. Prolonged heating at high temperature results in the formation of an insoluble chromium sulfate precipitate leading to bumping difficulties during distillation. Remove the flask from the ring stand and cool it first under the hot-water tap and then under the cold. Wash the thermometer and remove it. Add 100 cc. of distilled water cautiously while rotating the flask. Connect the flask with the distillation apparatus. To a 250-cc. flask containing an antibump, add 2-cc. of *N* potassium carbonate and place the flask under the condenser stem in such a manner that the tip of the stem dips into the liquid. Place 10 cc. of phosphorous acid 50% in the reservoir on the top of the apparatus and after bringing the liquid to a boil so as to wet the glass joints, allow the acid to run in slowly by turning the stop-cock. Rapidly distil over 75 cc. Lower the flask containing the distillate, open the stop-cock, and wash down the condenser stem permitting the washings to join the distillate. Remove the flask to a burner and boil down to a volume of 4.5 to 5.0 cc. Wash the antibump and remove it. Place the flask on a steam-bath and add from a dropper enough 0.2*M* potassium permanganate to establish a permanent wine-red coloration. Heat five minutes with frequent rotation. Add 10 drops of syrupy phosphoric acid and continue to heat for five minutes. No decoloration of the permanganate