chloroform, treated with activated charcoal and filtered through a charcoal mat on a Buchner funnel. The filtrate was evaporated to dryness and extracted with ether to remove impurities. The residue not dissolved by the ether was taken up in chloroform and the solution washed with several small portions of water. The chloroform was removed *in vacuo*, the residue obtained was dissolved in a known quantity of alcohol. Two volumes of ether were then added, the mixture shaken out with water and the aqueous layer filtered into a small dish. This was placed in a vacuum desiccator and evaporated to complete dryness. The transparent residue possessed the following properties:

1. It was tan in color.
2. It dissolved readily in water, alcohol and chloroform, but not in ether.
3. When hydrolyzed by boiling with dilute sulfuric acid, it reduced Fehling's solution. The untreated residue had no reducing power.
4. The solid and its solution possessed a bitter taste.
5. Mayer's reagent gave a negative text.
6. Tannic acid produced a copious white precipitate.
7. It showed the following actions on being heated:

Softening	94–100° C.
Fusion	107–108° C.
Swelled	120° C.
Darkened	160–180° C.
Reddish brown	203–205° C.

The heated mass was dissolved in water, and the solution tested. The substance no longer showed the properties of a carbohydrate.

8. After standing for eight days in a stoppered vial, it exhibited the same properties as before and was found to be toxic to sparrows.

The yield of the glycoside was approximately 0.002%.

In several other experiments, the glycoside was precipitated by addition of tannic acid solution to a purified extract of the rhizome. The precipitate was collected and treated with litharge. The dried mass was then extracted with absolute alcohol but no glycoside was obtained. Hence, it is apparent that this method should be used with caution.

CONCLUSIONS

1. Sucrose and glucose were present in the aqueous and alcoholic extracts obtained from parts of *A. Cornuti*.

2. The leaves and stems contain a bitter principle which may be divided into a toxic and a non-toxic fraction.

3. The leaves and stems examined did not contain any significant amount of glycosides.

4. A toxic glycoside which resembled asclepiadin in properties was isolated from the rhizome of *A. Cornuti*, apparently in impure form.

5. The overground portions of *A. Cornuti* contained a principle which was irritating to the dermis.

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Studies on Cantharides. I. The Titration of Cantharidin*,†

By Benjamin P. Hecht and Lloyd M. Parks‡

INTRODUCTION

For many centuries the drug Cantharides, commonly referred to as Spanish or Russian Flies, has been used in medicine. Its history dates at least as far back as the time of the early Greeks, as shown in the writings of Hippocrates.

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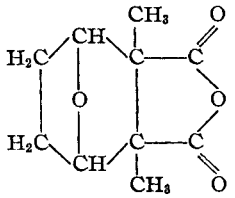
In medicine, the principle action of Cantharides is that of a vesicatory and the various uses to which it has been applied depend, more or less, upon this effect. The therapeutic action of the drug has been shown to depend upon the chief constituent, cantharidin, which was first isolated in 1810 by Robiquet (1). The chemistry of this substance has been developed by Picard (2), Homolka (3), Anderlini (4), Spiegel (5), Meyer (6), Danckwortt (7), Gadamer (8) and Rudolph (9). The following accepted structure has been advanced for cantharidin:



Since the efficacy of the drug depends entirely upon this active principle, it has been necessary to determine the content of cantharidin in evaluating Cantharides. Unfortunately, the methods that have been developed thus far have not proved to be satisfactory. These methods have all depended upon the separation of cantharidin in pure form and weighing the amount thus obtained. The present assay process of the U. S. Pharmacopœia, Eleventh Revision, which is of such nature, has several disadvantages. It gives results which are not accurate, for in the final step of the process, the cantharidin is not obtained in the pure state, but is always associated with inert material which is not easily removed. The process is also unusually long and time consuming.

Since cantharidin has been shown by Homolka (3), Danckwortt (7) and Gadamer (8) to be the anhydride of a dibasic acid, it seemed possible and practicable to determine it by acidimetric titration. The analysis of the di-methyl ester of cantharidin by Homolka, and of the potassium salt by Danckwortt, indicated a dibasic anhydride structure, the neutralization of which takes place according to the following equation:

$$C_{10}H_{12}O_4 + 2M'OH = C_{10}H_{12}O_5M_2' + H_2O$$

Upon reviewing the literature, it was found that cantharidin could not be titrated quantitatively. In this connection, Scoville (10), (11) has published the results of his unsuccessful attempts to titrate cantharidin acidimetrically, from which he concluded that the compound does not combine with alkali in any definite proportion during titration.

The discrepancy involved in the titration of cantharidin has been explained by Gadamer (12). He believed it to be due to hydrolysis of the salt formed, and has laid down the following system:

$$C_{10}H_{12}O_5K_2 + 2\ H_2O = C_{10}H_{12}O_5H_2 + 2\ KOH$$
$$KOH = K^+ + OH^-$$

Accordingly, the presence of excess hydroxyl ions account for an end-point before all of the anhydride can be titrated. The same explanation has been advanced by Danckwortt.

Danckwortt (7) has observed that about $1^1/_2$ carboxy groups can be accounted for in the titration of cantharidin. It is significant to note that Mulliken (14) does not include a neutralization equivalent for cantharidin.

From the above, it may be seen that any approach to a titrimetric method for the estimation of cantharidin in Cantharides must first be concerned with the titration of pure cantharidin itself. Once this is accomplished satisfactorily, the results may be applied to the drug and a satisfactory assay process may be developed.

EXPERIMENTAL

Inasmuch as it had been concluded from the work of Danckwortt (7), Scoville, (10), (11) and Gadamer (12) that cantharidin cannot be titrated quantitatively, it seemed desirable to repeat some of their experiments.

A quantity of cantharidin was obtained from S. B. Penick and Company. It had a melting point of 210–212° (uncor.), appeared in granular form and on careful examination exhibited a yellow color. The product was therefore purified by recrystallizing it from hot chloroform. Colorless, tabular prismatic and hexagonal-shaped crystals were obtained, M. P. 214–214.5° (uncor.).

DIRECT TITRATION OF CANTHARIDIN

In carrying out titrations of cantharidin, it was necessary to employ a solvent such as alcohol or acetone because of the insolubility of this compound in water and even in solutions of alkali, unless heated for some time.

With Aqueous Potassium Hydroxide.—A 0.1500-Gm. sample in neutral acetone solution gave no definite end-point when titrated with aqueous alkali, using phenolphthalein indicator. Addition of the aqueous alkali precipitated the cantharidin from acetone solution.

With Alcoholic Potassium Hydroxide.—To prevent the precipitation of cantharidin, alcoholic potassium hydroxide was used and the above experiment repeated. 0.1000 Gm. of cantharidin required about 6.4 cc. of $\frac{N}{10}$ alcoholic KOH, as compared to 10.2 cc., theoretical. The end-point was very indefinite.

RESIDUAL TITRATION OF CANTHARIDIN

In Acetone Solution.—Cantharidin was dissolved in neutral acetone, an excess of $\frac{N}{10}$ alcoholic KOH added, and the solution then titrated to neutrality with $\frac{N}{10}$ HCl, using phenolphthalein as indicator.

Sample	Gm. Cantharidin	Cc. $\frac{N}{10}$ KOH Required	Cc. $\frac{N}{10}$ KOH Calc.	% Deviation
1	0.1000	6.85	10.2	32.84
2	0.1000	8.6	10.2	15.68

This method gave a very sharp end-point, but the results were low and differed greatly in the two titrations.

In Alcoholic Solution.—The above experiment was repeated, with the cantharidin in alcohol solution.

Sample	Gm. Cantharidin	Cc. $\frac{N}{10}$ KOH Required	Cc. $\frac{N}{10}$ KOH Calc.	% Deviation
1	0.1000	6.8	10.2	33.35
2	0.1000	6.4	10.2	37.25

Again, the end-point in these titrations was sharp but the results remained low. These low results suggested that neutralization was incomplete, possibly because of the resistance which the anhydride structure offered to the action of alkali.

RESIDUAL TITRATION OF CANTHARIDIN AFTER REFLUXING WITH ALKALI

With Alcoholic Alkali.—In order to overcome the resistance of the anhydride structure to the action of alkali, which was suspected from the above experiments, heat was applied.

0.1000 Gm. of cantharidin was dissolved in 25 cc. of neutral acetone, 25 cc. of $\frac{N}{10}$ alcoholic KOH added, and the solution refluxed for $^1/_2$ hour. The solution was then cooled, and the excess alkali titrated with $\frac{N}{10}$ HCl, using phenolphthalein as indicator.

0.1000 Gm. cantharidin required 6.5 cc. of $\frac{N}{10}$ KOH as compared to 10.2 cc., theoretical. Per cent deviation, 36.27.

With Aqueous Alkali.—Cantharidin was refluxed with an excess of $\frac{N}{10}$ aqueous KOH until solution was effected and the excess alkali then titrated with $\frac{N}{10}$ HCl, using phenolphthalein as indicator.

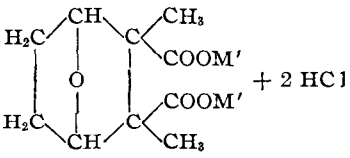
Sample	Gm. Cantharidin	Cc. $\frac{N}{10}$ KOH Required	Cc. $\frac{N}{10}$ KOH Calc.	% Error
1	0.1684	17.4	17.18	1.27
2	0.1751	17.8	17.86	0.33
3	0.2153	22.1	21.96	0.47

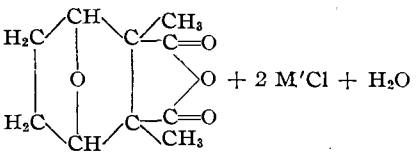
It required about an hour's refluxing before the solution of cantharidin resulted, the time varying with the fineness of the cantharidin.

The results of the first of these experiments verified the work of Scoville (10). The results of the latter experiment, however, showed that cantharidin can be titrated quantitatively in the absence of organic solvents, and indicated that the presence of such solvents, or, perhaps, the absence of water, are factors involved in the neutralization of the anhydride structure by alkali.

RESIDUAL TITRATION OF CANTHARIDIN, USING A WEAK ACID

It is well known that the addition of mineral acids to salts of cantharidic acid, the dibasic acid resulting upon the hydration of the acid anhydride, causes decomposition of these salts with the formation of cantharidin, the dibasic acid being quite unstable:





That this might explain the failure to titrate cantharidin was held plausible. Accordingly, a residual titration was tried, in which hydrochloric acid was replaced by a weak acid, potassium acid phthalate. A 0.1000-Gm. sample required 9.0 cc. of $\frac{N}{10}$ KOH as compared to 10.2 cc. theoretical. This low value indicated that decomposition of the salt does not occur during the residual titration of cantharidin when using strong mineral acids.

EFFECT OF STRONG ACIDS ON SALTS OF CANTHARIDIC ACID

There was the alternate possibility, in the above experiment, that potassium acid phthalate, a weak acid in the ordinary sense, was strong enough, on the other hand, to decompose the salts of cantharidic acid formed during titration. To investigate this possibility new experiments were devised to determine the effect of strong acids on the salts of cantharidic acid. The behavior of cantharidates toward strong acids is essentially that of a buffer action. If these salts performed as buffers during residual titrations, the low values experienced would become apparent.

Barium cantharidate and potassium cantharidate were prepared by neutralizing cantharidin with hot solutions of barium hydroxide and potassium hydroxide, respectively, and purifying the resulting salts. To 10 cc. of $\frac{N}{10}$ $Ba(OH)_2$ a quantity of barium cantharidate was added and the mixture titrated to neutrality with $\frac{N}{10}$ HCl, using methyl orange. The experiment was repeated with the addition of potassium cantharidate to 10 cc. of $\frac{N}{10}$ KOH. In each case 10 cc. of $\frac{N}{10}$ HCl were required for the titrations.

In these titrations the amount of acid required was equivalent in each instance to the amount of alkali originally used. This demonstrated that the presence of salts of cantharidic acid, formed during the residual titration of cantharidin, does not interfere with the process. Therefore, the buffer action of cantharidates is not a factor and does not explain the low results obtained in the titrations thus far. These experiments also showed that a strong mineral acid, such as hydrochloric, may be employed in residual titrations of cantharidin.

ATTEMPT AT RESIDUAL TITRATION OF CANTHARIDIN, USING BARIUM HYDROXIDE

It appeared thus far that the explanation advanced by Gadamer (12) (hydrolysis of the salt formed) to explain the failure to titrate cantharidin quantitatively, might have some foundation. But if this were the case, the neutralization could be made to go to completion by using barium hydroxide as the base. Since barium cantharidate is insoluble in water, the reversible reaction, and hydrolysis of the salt, would be prevented in accordance with the Law of Mass Action. Accordingly, this possibility was thought worthy of investigation. It was attempted to use barium hydroxide in the residual titration of cantharidin in alcohol solution, using a hot plate to keep the substances in solution. However, it was impossible to obtain a definite end-point and further work using barium hydroxide as the alkali was abandoned.

DETERMINATION OF THE p_H OF POTASSIUM CANTHARIDATE SOLUTIONS

A number of aqueous solutions of potassium cantharidate of various molar strengths were prepared and the p_H of these determined by means of a Cameron p_H Meter, using a glass electrode. The readings were made at a temperature of 25°. The results were as follows:

Strength of Potassium Cantharidate	p_H
$M/20$	10.25
$M/200$	10.20
$M/400$	10.00
$M/800$	9.85
$M/1600$	9.60
$M/2000$	9.58

By determining these p_H values several facts were revealed which ultimately led to a better understanding of the problem.

First, phenolphthalein was shown to be a correct indicator to use in the titration of cantharidin when a strong base is employed, as the salt had a p_H of 10 ($M/400$). Thymolphthalein is also a suitable indicator, and perhaps to be preferred.

Indicator	p_H Range	Transition
Phenolphthalein (0.003M)	8.3–10	9.3
Thymolphthalein	9.5–10.5	9.5

Second, some idea as to the extent of hydrolysis was obtained. Because of certain properties of cantharidin the experimental determination of the degree of hydrolysis was made difficult. In carrying out such a measurement, it was first necessary to determine the dissociation constant of cantharidic acid, the hydrate of cantharidin. This acid is, however, unstable and cannot be isolated, the addition of strong acids to solutions of the salt causing decomposition with the precipitation of cantharidin. However, by finding the p_H of potassium cantharidate, an approximation of the dissociation constant of this acid was obtained.

Relatively, cantharidic acid was shown to be stronger than boric acid but weaker than hydrosulfuric acid. This was demonstrated by adding acids of different dissociation constants to a solution of potassium cantharidate, and observing if precipitation of cantharidin occurred.

Relative Strength of Cantharidic Acid

Acid Added to Solution of Potassium Cantharidate	Dissociation Constant of Ac	Precipitation of Cantharidin Observed
Hydrochloric	.....	+
Acetic	1.86×10^{-5} 25°	+
Propionic	1.4×10^{-5} 25°	+
Butyric	1.53×10^{-5} 25°	+
Caproic	1.43×10^{-5} 18°	+
Hydrosulfuric	5.7×10^{-8} 18°	+
Cantharidic	.....?	..
Boric	6.6×10^{-10} 25°	–

The first five acids were added directly to the solution of the salt; hydrosulfuric was added by passing H_2S through the solution for several hours; boric acid was added by first dissolving it in distilled water.

From the p_H data, a more definite value for the dissociation constant of cantharidic acid was obtained. The $M/200$ solution of potassium cantharidate, corresponding to 0.01 normal, had a p_H of 10.2. According to the data of Bjerrum (13), this value lies between 4 and 3 ($H^{-14} - H^{-10.2} = H^{-3.8}$). By interpolation, the dissociation constant of cantharidic was calculated to be $10^{-3.4}$, in round numbers. In the same manner a p_H of 9.58, found for the $M/2000$ solution, corresponded to $10^{-3.2}$ as the dissociation constant. Taking the mean of these two values, the dissociation constant of cantharidic acid was placed at $10^{-3.3}$, equivalent to 5×10^{-9}.

It was now possible to calculate the per cent of hydrolysis of the salt, since there exists a mathematical relationship between hydrolysis of the salt and the dissociation constant of the acid. Since the latter value was found to be greater than 10^{-6}, the degree of hydrolysis is more than 1% and the following mathematical expression is applicable:

$$\frac{CX^2}{1 - X} = \frac{Kw}{Ka} = Kh$$

where

C = molar concentration of the salt
Kw = ion product of water
Ka = dissociation constant of the acid
X = degree of hydrolysis

Substituting, using 1.27×10^{-14} as the value of the ion product of water at 25°,

$$\frac{0.005X^2}{1 - X} = \frac{1.27 \times 10^{-14}}{5 \times 10^{-9}}$$

solving,

$$X = 0.0228$$

The degree of hydrolysis of potassium cantharidate in $M/200$ concentration at 25° is therefore 2.28%. While this value found for X may not be entirely accurate, it is, nevertheless, a close approximation.

In the titration of cantharidin, any error involved due to hydrolysis of the salt should be in the order of 2%. By proper choice of an indicator, such as phenolphthalein, this error can be overcome or at least much reduced. Actually, the magnitude of error experienced was much greater, although the per cent of deviation varied considerably.

	Per Cent Deviation
1	33.35 (in alcohol)
2	37.25 (in alcohol)
3	32.84 (in acetone)
4	15.68 (in acetone)

RESIDUAL TITRATION OF CANTHARIDIN WITH REMOVAL OF ORGANIC SOLVENT

That the alcohol or acetone which had to be used as a solvent for cantharidin must have some effect on the titration results was strongly indicated by the results of previous experiments. These solvents have low dielectric constants and may greatly influence ionization. Since neutralization depends upon H^+ ions and OH^- ions furnished by acid and base, respectively, depression of the ionization of either would account for anomalies in titrations. In spite of the fact that alcohol affects the ionization of fatty acids, these acids can still be titrated in alcohol solution. With the idea in mind that the ionization of cantharidin is greatly depressed by these organic solvents, a method was devised whereby they could be removed.

Cantharidin was dissolved in 25 cc. of neutral acetone, followed by an excess of $N/10$ alcoholic KOH, and 25 cc. of distilled water. The solution was then evaporated until the acetone and alcohol were removed, leaving an aqueous solution of potassium cantharidate and an excess of potassium hydroxide. The excess alkali was then titrated with $N/10$ HCl, using phenolphthalein. A correction was applied by running a blank for the acetone. The following results were obtained:

Sample	Gm. Cantharidin	—Cc. $\frac{N}{10}$ KOH— Required	Calculated	% Error
1	0.0949	9.5	9.68	−1.85
2	0.0857	8.5	8.74	−2.74
3	0.0567	5.9	5.78	+2.07
4	0.0700	7.4	7.14	+2.77
5	0.0800	8.1	8.15	−0.61
6	0.1522	15.5	15.53	−0.19
7	0.2007	20.4	20.47	−0.34

By this method of titration the results were quantitative, within the limits of experimental error. The removal of the alcohol and acetone, which had to be used originally to dissolve the cantharidin, permitted complete neutralization. This showed fairly conclusively that the failure to titrate cantharidin by any of the methods hitherto used was due to a depression of the ionization of cantharidin by alcohol, acetone or benzene, or to the absence of water as a hydrating medium. As a result there was required a greater amount of H^+ ions which had to be supplied by the acid used in the residual titration, and this in turn caused the low results always experienced. Additional evidence that the presence of these organic solvents prevented the quantitative titration of cantharidin had already been furnished by the results of a previous experiment on the residual titration of cantharidin after refluxing with aqueous alkali, in which, in the absence of organic solvents, quantitative results were obtained.

STUDY OF THE TITRATION OF OTHER WATER INSOLUBLE ANHYDRIDES

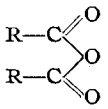
Inasmuch as cantharidin is an acid anhydride, insoluble in water, it was thought that other water-insoluble acid anhydrides should behave similarly upon titration. Accordingly, a study was undertaken to determine the effect of alcohol on the titration of two other water-insoluble acid anhydrides, benzoic and phthalic acid anhydrides.

RESIDUAL TITRATION OF BENZOIC AND PHTHALIC ANHYDRIDES IN ALCOHOL

Benzoic and phthalic acid anhydrides were each dissolved in alcohol, followed by an excess of $N/10$ alcoholic KOH. The excess alkali was titrated with $N/10$ HCl, using phenolphthalein. A blank was run for the alcohol.

Sample	Anhydride	Gm.	Cc. $\frac{N}{10}$ KOH Required	Cc. $\frac{N}{10}$ KOH Calculated	% Error
1	Phthalic	0.2049	22.2	27.65	19.53
2	Phthalic	0.4413	36.6	59.63	38.58
3	Benzoic	0.2019	11.7	17.85	34.45
4	Benzoic	0.3034	21.1	26.85	24.41

From the above data the low results obtained are comparable to those obtained in the similar titrations of cantharidin. Thus the depression of the ionization of cantharidin is not due to some property exclusively inherent in its structure, but is common to other water-insoluble acid anhydrides as well. These experiments demonstrate that water-insoluble acid anhydrides, in general, cannot be titrated in organic solvents, such as alcohol and acetone in the absence of water. Furthermore, the structure of acid anhydrides is such that it should be expected to undergo ionization with difficulty, since it does not contain replaceable hydrogen:



This is entirely compatible with the fact that in alcohol or a similar nonionized solvent, ionization is made even more difficult. Organic solvents unquestionably affect the indicators as well, but the extent to which this occurs is not comparable to the effect which it exerts on the water-insoluble acid anhydrides.

RESIDUAL TITRATION OF BENZOIC AND PHTHALIC ANHYDRIDES AFTER REFLUXING WITH AQUEOUS ALKALI

This experiment was carried out in a similar manner to that used for cantharidin in a previous experiment. The anhydrides were refluxed with an excess of $N/10$ aqueous KOH until solution was affected and the excess alkali then determined by titration with $N/10$ HCl, using phenolphthalein.

Sample	Anhydride	Gm.	Cc. $\frac{N}{10}$ KOH Required	Cc. $\frac{N}{10}$ KOH Calculated	% Error
1	Phthalic	0.3012	41.1	40.70	−1.47
2	Phthalic	0.2017	28.2	27.98	−0.71
3	Phthalic	0.3110	43.5	43.37	+0.29
4	Benzoic	0.2002	17.8	17.71	+0.56
5	Benzoic	0.2952	26.2	26.12	+0.35

These results, as in the case of cantharidin, are quantitative within the limits of experimental error, and again emphasize the point that water-insoluble acid anhydrides are titratable, but not in the exclusive presence of organic solvents.

RESIDUAL TITRATION OF BENZOIC AND PHTHALIC ANHYDRIDES WITH REMOVAL OF ORGANIC SOLVENT

The method used here was the same as that used for cantharidin in a previous experiment. The anhydride was dissolved in 25 cc. of alcohol, followed by an excess of $N/20$ alcoholic KOH, and 25 cc. of distilled water, and the solution evaporated to a volume of about 10 cc., or until the alcohol was no longer detectable. The excess alkali was then titrated with $N/10$ HCl, using phenolphthalein. A blank was run for the alcohol used.

Sample	Anhydride	Gm.	Cc. $\frac{N}{20}$ KOH Required	Cc. $\frac{N}{20}$ KOH Calculated	% Error
1	Phthalic	0.2906	78.1	78.54	−0.56
2	Phthalic	0.1681	45.6	45.43	+0.37
3	Benzoic	0.2008	34.8	35.52	+2.02
4	Benzoic	0.2004	35.6	35.46	+0.39

The results obtained here, as in the similar experiment, are again quantitative. This method of titrating water-insoluble acid anhydrides would be especially useful for determining the neutralization equivalents or for the quantitative estimation of those acid anhydrides which are water insoluble and which may be decomposed by the heat of refluxing, or are volatile.

SUMMARY AND CONCLUSIONS

1. The results of other investigators, namely, Scoville, Gadamer and Danckwortt, in their attempts to titrate cantharidin quantitatively in the presence of such organic solvents as alcohol, acetone or benzene, have been confirmed.

2. The failure to titrate cantharidin in the presence of organic solvents has been shown to be due, not to hydrolysis of the salt formed, as advanced by Gadamer, or to the buffer action of the salt formed, or to the action of strong mineral acids on the salt formed; it has been shown to be due to some effect exerted by the organic solvent used to

dissolve the cantharidin. This effect is apparently a depression of the ionization or neutralization of cantharidin, due possibly to the low dielectric constant of the solvent and its inability to ionize.

3. The effect of alcohol on other water-insoluble acid anhydrides has been investigated and it was shown that the two studied, benzoic and phthalic anhydrides, behaved similarly to cantharidin in attempts to titrate them quantitatively in the presence of alcohol. The effect of alcohol is thus apparently not restricted to cantharidin, but is general for all water-insoluble acid anhydrides.

4. A method has been developed for the quantitative titration of cantharidin, in which the organic solvent used to dissolve the compound is removed completely during the procedure, in the presence of water and excess alkali. This method yielded equally satisfactory results for cantharidin and the other anhydrides, benzoic and phthalic, and is generally applicable to all water-insoluble acid anhydrides.

5. The p_H's of solutions of potassium salts of cantharidic acid of various molar strengths have been determined. From these it was possible to estimate the ionization constant of the free acid, which was calculated as being 5×10^{-9}, stronger than boric acid but weaker than hydrosulfuric acid. It was also possible to calculate the degree of hydrolysis of the potassium salt as being approximately 2%.

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The Sterol and Resin Alcohols from Celastrus Scandens

*By Ole Gisvold**

During the investigation of the pigments contained in the petroleum ether extract of the outer bark of the root of *Celastrus Scandens*, it was deemed advisable to investigate the phytosterols which might be present. In addition to the sterol that was apparently a sitosterol or a mixture of sitosterols, three resin alcohols were isolated.

EXPERIMENTAL

Phytosterol.—The pigment-free petroleum ether extract obtained from the outer bark of the root of *Celastrus Scandens* was saponified with alcoholic potassium hydroxide and the nonsaponifiable residue extracted by means of a continuous extraction apparatus with Skelly-solve B. The solvent was removed on a steam-bath and the residue taken up in alcohol. Fractional crystallization enabled the separation of some phytosterol. The residual sterol was isolated by means of its digitonide (1). The digitonide was decomposed by precipitating the digitonin with ether from a pyridine solution of the complex (2).

The purified sterol melted at 137.5° C. and its acetate at 121° C. It had a specific rotation of −32.5 in chloroform at 25° C. It gave the color reactions characteristic of sterols.

A number of sterols have been reported in the literature having physical constants closely approaching those given above. It is highly possible that it is a mixture. The amount isolated was too small to warrant further investigation.

Resin Alcohol No. 1.—The sterol-free nonsaponifiable material upon spontaneous concentration deposited needle-like crystals in an oily residue. The oily material was successfully removed by washing with cold Skelly-solve B. The crystals purified by several recrystallizations from Skelly-solve B became soft at 215° C. and clear at 221° C.

These crystals gave a deep purple color when subjected to the Lieberman-Burchard test. The material available was too small for further investigation.

Resin Alcohol No. 2.—The mother liquor upon spontaneous evaporation left a very viscous oil from which needle-like crystals slowly separated.

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