A CHEMICAL EXAMINATION OF THE ROOT OF LEPTOTÆMIA DISSECTA.*

BY NELLIE WAKEMAN.

In April 1923 there were received from Dr. E. T. Krebs of the Balsama Company of San Francisco, California, 100 lbs. of root of *Leptotæmia Dissecta* collected during the previous summer in the high Sierras. According to Dr. Krebs' letter the plant from which the root was collected has been identified by Professor Smiley of the University of California. (Letter to Professor Kremers under date of April 13, 1923.) "Other botanists," the writer adds, "classified it differently, but probably due to specimens reaching them in poor condition."

The following additional information taken from the letter referred to above should prove of interest:

"The plant grows on the eastern side of the Sierras at an elevation of 5000 feet or more and only on the east and south slopes. It comes up in March, blooms in May and dies in August of the average year. We have found it as far as the Oregon line north and as far south as Owens Lake in Inyo County. It is the sacred plant of the Washoe Indians who formerly ran over this area, and is called by them *Dortza*. The roots shoot up one stalk the first year and each succeeding year another stalk. Therefore, a plant with eight or ten stalks or knobs on the top of the tuber is estimated to be that many years old. It grows under grease wood, sage brush or other high brush, as a rule, and if the Leptotæmia root is near the root of the brush the latter dies.

"It might interest you as preliminary information to know that I have separated from the root the following: sugars, gums, resins, fixed oil, volatile oil, a neutral principle and an acid. No alkaloids are present so far as I could determine. It contains a large amount of sugars. The gum is about twenty per cent. by weight of the dried root. The fixed oil is also in large amount. I am interested if it contains any natural esters."

All of the ground root, 100 pounds, was placed in the 60-liter copper still of the station which is operated with steam under pressure from the Central Heating Plant. The original oil weighed 178 grams. The large volume of aqueous distillate was cohobated yielding 9 grams of cohobated oil, a total of about 0.42 p. c. By distilling the alcoholic extract of the root 35 grams more of volatile oil were obtained and by cohobating the aqueous distillate, another 40 grams, making in all 274 grams of volatile oil, a little less than 0.6 p. c.

For both the original oil and the cohobated oil the ordinary physical constants were taken. The cohobated oil was dried over calcium chloride on account of its turbidity.

	Original oil.	Cohobated oil.
Density	0.936 (at 24°)	0.917 (at 20°)
Angle of rotation	−4.16 (at 25°)	−1.90 (at 23°)

It is worthy of note that the density of the cohobated oil is less than that of the original oil.

After distilling off the volatile oil the root was thoroughly extracted with alcohol and the alcohol evaporated. About 25 liters of extract remained. This separated into two layers, a thick, black oily portion and a dark colored aqueous portion.

* Scientific Section, A. Ph. A., Buffalo meeting, 1924.

All of the foregoing work was carried out by Dr. R. E. Kremers and Carl Kremers during the spring and summer of 1923. In October 1923 the material was turned over to the writer.

THE VOLATILE OIL.

The physical constants of the oil remained unchanged. The oil was slightly acid to litmus paper but showed practically no diminution when 10 cc. were shaken in a cassia flask with 5 p. c. sodium hydroxide solution. Phenols are therefore absent. It gave no reaction for aldehydes when treated with Schiff's reagent. Saponification with alcoholic potash showed an ester content of 14–15 p. c., calculated as menthyl acetate, or about 18 p. c. calculated as menthyl valerianate, in the original oil, and in the cohobated oil 20–22 p. c., calculated as menthyl acetate or 24–25 p. c., calculated as menthyl valerianate. Acetylization and subsequent hydrolysis showed total alcohols to equal 34 p. c. in the original oil, calculated as menthol, and 49–50 p. c. in the cohobated oil.

The original oil was now hydrolyzed by the use of potassium hydroxide in alcohol, the alcohol was evaporated and the oil distilled with steam; 82 p. c. of the oil being recovered.

The alkaline aqueous residue was made acid with sulphuric acid and the volatile acid thus liberated was distilled off. Both barium and zinc salts of this acid were prepared. The barium salt was slow to crystallize, but finally good crystals of both salts were obtained. The acid radicle of the former was determined by precipitating the barium as barium sulphate, also by double decomposition with silver nitrate and subsequent silver determinations. The zinc in the zinc salt was determined as both oxalate and carbonate. All the results indicated valeric acid. During the distillation and the concentration of the salt solutions a marked celery-like odor was noted.

The saponified oil, 135 cc., was distilled under ordinary pressure. Fractions were collected as follows:

150 degrees–200 degrees	26 cc.
200 degrees–230 degrees	19 cc.
230 degrees–250 degrees	25 cc.
Above 250 degrees (Residue)	62 cc.

The first three fractions were practically colorless, but when 250° was reached the oil began to turn yellow, indicating decomposition. The heat was therefore discontinued.

These fractions were again refractionated under diminished pressure, beginning at 25 mm. and working down to 7 mm. pressure. No fraction of more than 25 cc. was obtained within a temperature range of 10°. The specific gravity of the fractions increased gradually from 0.86 in the lowest, to 0.95 in the highest boiling fraction.

While redistilling the original 2nd fraction (200° to 230° under ordinary pressure) a solid white substance, resembling borneol in appearance, separated in the condenser. This was collected in the fractions boiling at 120° to 130°, and 130° to 140°, under 25 mm. pressure. These fractions were tested for borneol by the bornyl phenylurethane method, according to Bertram and

Walbaum (*Jr. f. prakt. Chem.*, 49, p. 5). No bornyl phenylurethane was obtained. Repeated attempts to form crystalline derivatives of alcohols, from these and other fractions, met with negative results, though identification by means of benzoic and phthalic acid esters and phenyl- and naphthyl-urethanes was tried several times.

THE ALCOHOLIC EXTRACT.

The large volume of alcoholic extract, from which the alcohol had evaporated, formed two distinct layers. These were separated by means of a separatory funnel. About $4^1/_2$ liters of a dark oily portion were obtained and, perhaps, 5 times that volume of an aqueous portion. Both portions were strongly acid to litmus.

Acid and saponification values of the oily portion were determined with the following results:

	Sample I.	Sample II.
Acid value	108.5	115.0
Saponification value	220.8	218.0

The oily portion was now placed in the large still and distilled with steam. About 35 cc. of volatile oil with a marked odor recalling both chloroform and petroleum ether and a large volume of acid aqueous distillate were collected. It was afterwards learned that both chloroform and xylene had been added to the extract to preserve it after the alcohol had evaporated off. The oil was therefore contaminated with these substances.

Acid and saponification numbers of the nonvolatile portion of the oily extract were now obtained. These are here tabulated, for purpose of comparison, with those obtained before distilling off the volatile portion.

	Before distillation.		After distillation.	
	Sample I.	Sample II.	Sample I.	Sample II.
Acid value	108.5	115.0	125.0	126.9
Saponification value	220.8	218.0	161.8	169.5
Ester value	112.3	103.0	36.8	43.1

From the above it will be seen that while the total acid value of the oily extract had considerably diminished, it had been at the expense of the esters, while the free acid content of the extract had been increased.

The acid distillate was now neutralized with sodium carbonate, then cohobated and recohobated, until it was reduced to about 2 liters. By this process another 40 cc. of oil was obtained. The specific gravity of this oil was 0.955 at 20 degrees. Its odor recalled geraniol. Ester and alcohol determinations gave 24 to 25 p. c. of the former, calculated as menthyl valerianate, and 49 to 50 p. c. total alcohols, calculated as menthol.

The aqueous distillate (2 liters) was distilled fractionally. It began to boil at about 60 degrees. Fractions were collected as follows:

60–70°; 70–80°; 80–95°; 95–99°.

None of the fractions responded to tests for aldehydes. The three lower fractions gave tests for ethyl alcohol and for methyl alcohol. The ethyl alcohol test means nothing, however, since that alcohol had been used for extracting the

drug. An odor suggesting nitrogen bases was noticed while distilling, therefore the fraction boiling between 95 degrees and 99 degrees was collected in hydrochloric acid. The crystalline hydrochloride was obtained by evaporation. This was dissolved in alcohol and precipitated by acid platinic chloride solution. A bright yellow precipitate was obtained. This was found to contain 41.9 p. c. of platinum. By computation, the platinum content of the platinic chloride double salt of methylamine hydrochloride, $(CH_3NH_3)_2PtCl_6$, was found to be 41.3 p. c. The basic substance is therefore, undoubtedly, methylamine.

The sodium salt of the acid, remaining in the still, was crystallized and the acid was determined as before through the silver and zinc salts. Like the acid in the volatile oil, it was found to be valeric acid. Here, again, in evaporating the salt solutions to crystallization a strong celery-like odor was noticed.

SUMMARY.

The root of *Leptotæmia dissecta* contains about 0.6 p. c. of a volatile oil of high boiling point and high specific gravity. The oil is devoid of aldehydes and phenols, but contains 20–30 p. c. of the valeric acid ester of an as yet unidentified alcohol, and about as much free alcohol. From the fraction of saponified oil boiling between 200 degrees and 230 degrees a white crystalline substance separated while redistilling under diminished pressure.

The alcoholic extract of the drug separated, after the evaporation of the solvent, into an aqueous and an oily layer, both acid in reaction. The oily layer contains much acid and ester. In the aqueous distillate from this oily portion valeric acid, methyl alcohol and methylamine have been identified.

Further work is in progress, especially upon the oily and the aqueous extracts, of which a considerable quantity remains. This will be reported upon later.

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THE VOLATILE OIL OF MENTHA CANADENSIS L.*

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The statement which is frequently made that the Japanese menthol-producing peppermint is a variety of the species *Mentha arvensis*, makes it very desirable to investigate the native American mints of the *arvensis* and *canadensis* species. It is to be expected that such studies will not only help to clear up the relationships existing between the various plants, but that they ought also to throw light on the general mechanism of oil production.

Through the kind coöperation of Mr. O. A. Beath of the Wyoming Experiment Station, Laramie, a large quantity of *Mentha candensis* herb was received for investigation. The material was collected in Plumbago Canyon, Albany County, Wyo., August 20–23, 1922, at an altitude of 7000 feet. The plants were mostly in bloom

* Contribution from the Wisconsin Pharmaceutical Experiment Station. Studies in the Genus *Mentha V.*

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