

## Note on a micromethod for the estimation of ascaridole

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A micro stoichiometric method for the determination of ascaridole in a pure state and in oil of chenopodium is described. The determination is made either by direct or indirect titration of stannous chloride solution. A blank standardisation for the stannous chloride solution should be carried out consecutively.

**A** SCARIDOLE (1,4-epidioxy-2-*p*-menthene) is the main constituent of oil of chenopodium. Methods in the American (XII), British (1953) and Egyptian (1953) Pharmacopoeias are based on the fact that ascaridole liberates iodine from potassium iodide in concentrated acid medium. However, this method is empirically rather than stoichiometrically based.

In the last 25 years, several attempts have been made to modify the original Cocking and Hymas method. Lepetit (1943) stated that the factor in the official method (1 ml of 0.1 N sodium thiosulphate is equivalent to 0.00665 g ascaridole) should be reduced to 0.00605 g.

Böhme & Van Emster (1951) reported the titre to be affected by the concentration of ascaridole in the sample. Beckett, Donbrow & Jolliffe (1955) confirmed both Lepetit's and Böhme's findings. They gave a quadratic equation from which the amount of ascaridole can be calculated.

Said, Amer, Wahba & Ahmed (1962) described a method based on hydriodic acid as a reductant with heating. It was thought that it would be of interest to establish a simple and quick method for the evaluation of microquantities of ascaridole.

### MATERIALS

*Ascaridole* (Bayer) was purified by partition chromatography on silica column, using *n*-hexane-ethyl acetate mixture as the eluent (Beckett & Jolliffe, 1955). Purity was confirmed by physicochemical methods. B.p. at 20 mm/Hg, 112°;  $d_4^{20} = 0.9980$ ;  $n_D^{20}$ , 1.4769;  $[\alpha_D^{20}] \pm 0.00$ ;  $d_{20}^{20} = 1.002$ .

*Oil of chenopodium* (Rodofran Corporation) labelled to contain not less than 65% ascaridole, complied with the U.S.P. XII requirements.

*Stannous chloride* solution (1.5%): dissolve 1.5 g stannous chloride with heat in 50 ml concentrated hydrochloric acid, cool, adjust to 100 ml with freshly distilled water. The reagent should be recently prepared and kept under nitrogen.

*Ascaridole solution*, (0.5 and 5.0% w/v) in 90% acetic acid.

*Iodine solution* 0.05 N. The solution is stored in small air-tight bottles filled completely and kept in a cool, dark place.

*Starch test solution* as indicator.

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## MICROMETHOD FOR ASCARIDOLE

### Experimental

The proposed method is based on the fact that ascaridole can be estimated by its reaction with a powerful reducing agent. Ascorbic acid, sodium sulphite, hydriodic acid in the cold, and stannous chloride were tried. The latter proved to be the most favourable reducing agent in determining ascaridole in its pure state and in oil of chenopodium. The method of analysis can be made either by direct or indirect titration.

*Indirect titration.* Into a stoppered test tube, of about 25 ml capacity, 5 ml of stannous chloride solution is measured accurately. From a burette, 1–5 ml of ascaridole solution is added, followed by three drops of starch solution. The mixture is shaken and titrated immediately with the standard iodine solution until the appearance of the blue colour. A blank is estimated under the same conditions.

*Direct titration.* Into a stoppered test tube, of about 25 ml capacity, 5 ml of stannous chloride solution is measured accurately. Three drops of starch solution and one drop of 10% potassium iodide solution is added and the mixture titrated with the ascaridole solution until the appearance of permanent blue colour. Another 5 ml of stannous chloride solution is titrated with iodine using starch test solution as indicator.

TABLE 1. ANALYSIS OF DIFFERENT QUANTITIES OF ASCARIDOLE

Weight mg	ml 0.05 N iodine consumed†	mg ascaridole equiv. to 1 ml 0.05 N iodine
5	1.20	4.255
10	2.35	4.166
15	3.60	4.166
20	4.85	4.125
25	6.05	4.132

† Means of three estimations. Ascaridole equivalent to one ml of 0.05 N iodine solution is 4.1688 mg.

Analyses of different concentrations of ascaridole solution were made to find the reproducibility of the method. The results are presented in Table 1. It is clear that 1 ml of 0.05 N iodine is equivalent to 4.1688 mg ascaridole. As one molecule of ascaridole contains two atoms of oxygen which can be reduced, one ml of 0.05 N iodine is theoretically equivalent to 4.2057 or 8.4114 mg of ascaridole. The experimental findings are in favour of the first figure (4.2057).

A comparison between official and the stannous chloride methods was made. Two concentrations (0.5 and 5% w/v in 90 % acetic acid) of pure ascaridole and of oil of chenopodium were analysed. The results are shown in Table 2.

### Conclusions

The Pharmacopoeial methods for determining ascaridole involve the use of an empirical factor. The results we obtained were erroneous, especially at low concentrations. One molecule of ascaridole is equivalent to 4 atoms of iodine, indicating that the method is a stoichiometric

TABLE 2. COMPARISON BETWEEN THE OFFICIAL METHODS AND STANNOUS CHLORIDE METHOD

Ascaridole mg	Conc. of soln %	Found %*					
		Ascaridole			Oil of chenopodium		
		Official	Stannous chloride		Official	Stannous chloride	
			1.5%	5%		1.5%	5%
10	0.5	290.90	98.83	—	183.42	63.69	—
15		241.25	100.93	—	145.70	64.40	—
20		210.45	101.98	—	132.50	64.05	—
25		184.50	101.77	—	115.63	64.84	—
100	5.0	119.00	69.8	100.51	75.60	—	63.84
150		116.15	76.6	100.79	72.24	—	63.70
200		111.00	85.0	99.88	69.30	—	64.47
250		104.70	79.4	100.19	66.05	—	63.84

The indirect method of estimation was used for the comparison.

\* Means of three estimations.

On estimating high concentrations, low results were obtained in spite of using 5 ml stannous chloride solution per ml 5% ascaridole solution. However, on increasing the concentration of stannous chloride to 5% the expected values were obtained.

one. Our findings agree with those of Böhme & Van Emster and Beckett & others (1955) that the titres in the official methods are affected by the concentration of ascaridole (Table 2). The proposed method will estimate as little as 5–25 mg ascaridole, it is simple, requires no special apparatus and takes a comparatively short time.

## References

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