

## CORESTA RECOMMENDED METHOD N° 1

### DETERMINATION OF DITHIOCARBAMATES IN TOBACCO

*(November 1978)*

#### 1. PRINCIPLE

The dithiocarbamates are decomposed on heating with hydrochloric acid in the presence of stannous chloride. The carbon disulfide (CS<sub>2</sub>) which is formed, is transferred with a current of nitrogen, into a trap containing concentrated sulphuric acid to remove interfering substances, and then into a trap containing a methanolic solution of potassium hydroxide. The concentration of potassium *o*-methyl dithiocarbonate formed under these conditions is measured by spectrophotometry. As it is not normally known which dithiocarbamate is present in tobacco, results are expressed as carbon disulphide, and the values obtained using this method are taken to indicate the dithiocarbamate residue level.

#### 2. REAGENTS

1. Hydrochloric acid aqueous solution : 75 ml concentrated hydrochloric acid (AR) is added to 150 ml distilled water.
2. Stannous chloride : solid SnCl<sub>2</sub> (AR).
3. Concentrated sulphuric acid : chemically pure or AR.
4. Potassium hydroxide reagent : 56 g potassium hydroxide (AR) is dissolved in 1 liter methanol (AR) and 50 ml water is added to this solution. If there is any sediment the reagents should be filtered, using fluted filter paper before it is used.
5. Sodium diethyl-dithiocarbamate (a) : sodium diethyl dithiocarbamate trihydrate (AR) is used for the calibration.

#### 3. APPARATUS ( see figure 1)

A 250 ml three neck flask A is fitted with a water cooled condenser B, length 30 cm, a 100 ml reservoir C equipped with a stopcock and a tube reaching to the bottom of the flask, and a gas inlet D, which also reaches to the bottom of the flask. Both reservoir C and gas inlet D should be connected to a nitrogen supply, via a 3-way tap. The exit of the condenser is connected with two wash-bottles (E and F). The volume of each of the wash-bottles is about 80 ml and the inner tubes are equipped with GO sinters. The apparatus should be checked to ensure that there are no leaks.

#### 4. PROCEDURE

5 g of tobacco (b), weighed to the nearest 10 mg, is placed in flask A. 2 g stannous chloride is added followed by 50 ml distilled water.

The flask is shaken until all the tobacco has been impregnated. Immediately after this has been done, flask A is connected to condenser B, which is connected with wash-bottle E containing 20 ml concentrated sulphuric acid, and wash-bottle F containing 25 ml potassium hydroxide reagent. Reservoir C and inlet tube D are put in position, and a current of nitrogen, 50 ml per minute (c), is allowed to pass through the whole apparatus via D. Flask A is heated to 30-40 °C. As it is absolutely necessary that all of the tobacco is well impregnated by the stannous chloride solution, flask A is allowed to remain for at least 10 minutes in the conditions just described. This also has the advantage of purging any oxygen present in the apparatus.

Following the impregnation, 100 ml hydrochloric acid solution is placed in reservoir C and slowly added to flask A. Whilst the acid is being added to the reaction flask the 3-way tap should be turned so that the nitrogen supply is connected to reservoir C as well as passing into flask A via inlet tube D. This prevents any "suck-back" during the addition of the acid.

The contents of flask A are then heated to boiling point whilst maintaining a nitrogen flow of 50 ml per minute through inlet tube D. Boiling is sustained for 30 minutes. Condenser B must be well cooled to prevent steam water passing into the concentrated sulphuric acid in trap E.

At the end of the boiling period, wash-bottles E and F are disconnected and the nitrogen flow is turned off. The content of wash-bottle F is transferred to a 50 ml volumetric flask. Flask F is thoroughly rinsed with distilled water which is also added to the volumetric flask. The volume of the combined solutions is adjusted to 50 ml with distilled water. After mixing and allowing to stand for 15 minutes the spectrophotometric measurements are made at 272, 302 and 332 nm, using a 10 mm quartz cell, against a reagent blank of 25 ml potassium hydroxide reagent plus 25 ml distilled water (d). If any precipitation occurs in wash-bottle F, this indicates a high level of DTC, and the solution should be further diluted to 100 ml. Before every new analysis, the following procedure has to be observed :

1. The concentrated sulphuric acid in wash-bottle E must be renewed.
2. Wash-bottle E and F must be cleaned and dried, but acetone should not be used as any residue interferes with the subsequent analysis.

#### Remarks

- (a) Sodium diethyl-dithiocarbamate (Na DTC,  $3\text{H}_2\text{O}$ ) is the only pure dithiocarbamate which is readily available.
- (b) If possible, use a test sample that is in the form of cut tobacco or cigarette filler, without further preparation. If a test sample in these forms is not available, cut the laboratory sample into pieces of a suitable size. Grinding and drying lead to loss of dithiocarbamates.

- (c) The flow of nitrogen should be measured and pre-set before connecting to the apparatus.
- (d) The measured extinction at 302 nm shall not be over 0.800, nor under 0.100. If the extinction is over 0.800, a further dilution or a smaller amount of tobacco should be used. If the extinction is under 0.100, a quartz cell of longer path length should be used.

## 5. CALIBRATION

A solution of 59.2 µg/ml sodium diethyl-dithiocarbamate 3H<sub>2</sub>O, equivalent to 20 µg CS<sub>2</sub>/ml is prepared in water. This solution must be freshly prepared each day.

A range of standards, equivalent to 40, 60, 80, 100, 120 and 160 µg CS<sub>2</sub> is prepared by analysing 2, 3, 4, 5, 6 and 8 ml of this solution under conditions identical to those used for the analysis of tobacco.

A calibration curve is prepared by plotting amount of CS<sub>2</sub> in µg against extinction (ΔE), calculated using the following formula :

$$\Delta E = E_{302} - \frac{E_{272} + E_{332}}{2} \quad (1)$$

A calibration factor (f) may be calculated from the slope of the calibration graph :

$$f = \frac{\Delta E}{\mu\text{g CS}_2} \quad (2)$$

The calibration curve has been found to be very reproducible and a full curve need not be prepared each day. A single point check is normally sufficient.

## 6. CALCULATION

The amount of CS<sub>2</sub> in moisture free tobacco expressed in mg CS<sub>2</sub> per kg moisture free tobacco (p.p.m.) is :

$$\text{CS}_2 \text{ in mg/kg} = \frac{\Delta E \times 100}{f \times M \times (100 - W)}$$

ΔE = extinction, corrected as formula (1)

f = calibration factor calculated as formula (2)

M = tobacco weight (g)

W = moisture content of tobacco (%).

# FIGURE

RM 1 ter - Figure 1  
D.T.G. Apparatus

