

# Organic Application Note

## Fat in Cocoa Drink Mix

### Standard Method Used for Comparison

AOAC 963.15—Official International du Cacao et du Chocolat

### Accessories

501-081 Glass Wool; 502-327 LECO-Dry; 502-369 Kimwipe®; Isopropyl Alcohol

### Collection Vial Preparation Procedure

1. Cut 1.3 to 1.5 g of glass wool from the end of the glass wool rope.
2. Pull the compact section of glass wool apart so that the material is loosened considerably.
3. Pack the loosened glass wool into the collection vial with a clean spatula, a little at a time. The goal is to have random, not vertical orientation of wool strands.
4. Microwave the three collection vials together on high for three minutes in a 1000 watt household microwave. (The wattage rating of the microwave is usually found on the serial number sticker or serial number plate.) The time may be extended to four or five minutes for 750 watt microwaves.
5. Set aside vials and let them cool for 15 minutes before weighing.
6. Tare the empty balance pan.
7. Weigh collection vial and enter initial vial weight into the instrument.
8. Install the collection vial on the instrument collection system.

### Sample Preparation Procedure

1. Place 2.2 to 2.4 g of LECO-Dry into a 50 ml beaker.
2. Place beaker with LECO-Dry on balance and tare the weight.
3. Drop 0.9 to 1.1 g of cocoa mix onto the LECO-Dry.
4. Enter the sample weight into the instrument.
5. Remove the beaker from the balance. Thoroughly mix the cocoa with LECO-Dry, using a clean spatula.
6. Add 1.5 ml of isopropyl alcohol by pipette, onto the sample.
7. Thoroughly mix the sample, using a clean spatula. The prepared sample should not be sticky or adhere to the sides of the beaker. If this happens, reduce the amount of cocoa mix.
8. Install a lower end-cap assembly on a thimble and place in a thimble stand.
9. Pack 1/4 of a Kimwipe into the bottom of the thimble by folding it once and packing it into the bottom of the thimble with a clean spatula.
10. Transfer the prepared sample into the thimble using the funnel.
11. Install the upper end-cap assembly on the thimble.

### Extraction Parameters/Procedure

1. Set up (or recall and activate) the following instrument parameters:

|                        |            |
|------------------------|------------|
| Extraction Pressure    | 9000 psi   |
| Extraction Temperature | 100°C      |
| HVR Temperature        | 100°C      |
| Static Time            | 15 minutes |
| Dynamic Time           | 45 minutes |
| Flow Rate:             | 1.3 lpm    |



**TFE2000**

2. The pump head should be at 0°C or below from the last set of extractions. However, the refrigeration system times out and stops 20 minutes after extractions end. If the system has timed out, pre-cool the pump head by pressing any key on the key pad. The thimble and HVR temperatures should also be at set values. Temperatures can be displayed using the "Ambient Monitor" menu.
3. Insert the thimbles into the instrument and press the START key. The extraction will automatically take place, and the system will depressurize.

### Post-Extraction Manipulations

1. Remove the collection vials from the instrument.
2. Using the thimble removal tool, remove the thimbles and place them in the thimble stand to cool.
3. Microwave the three collection vials as done in step four of the collection vial preparation procedure.
4. Tare the empty balance pan.
5. Weigh each collection vial and enter the weight into the instrument.
6. Results will be automatically calculated. Choose the print option to receive a printout of the results.

### Typical Results (Total Fat by AOAC 963.15 = 15.2%)

| Sample ID | Weight (g)       | TFE2000 Fat (%) | Standard Method<br>AOAC 963.15 (%) |
|-----------|------------------|-----------------|------------------------------------|
| #1        | 1.0477           | 15.13           | 15.2                               |
|           | 0.9941           | 14.89           |                                    |
|           | 1.1139           | 15.32           |                                    |
|           | 1.0793           | 15.13           |                                    |
|           | 1.0093           | 15.60           |                                    |
|           | 1.0671           | 15.17           |                                    |
|           | <b>Average</b>   | <b>15.21</b>    |                                    |
|           | <b>Std. Dev.</b> | <b>0.24</b>     |                                    |

The LECO TFE2000 was developed to safely extract fats/oils from your food, feed, and oilseed samples by using an advanced analytical technique. Extensive research proves that liquid CO<sub>2</sub> at elevated pressures and temperatures is the most effective extraction solvent for these target applications. Extraction by CO<sub>2</sub> offers superior performance over conventional organic solvents plus:

**speed**—simultaneous extractions take just minutes to complete; method development is simplified

**low cost**—a CO<sub>2</sub> tank will supply extraction solvent for approximately 100 extractions; no disposal or recycle cost

**convenience**—no need for solvent evaporation steps in the method

**safety**—non-toxic, non-explosive



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