ASBC Methods of Hop Analysis

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Hop Bitterness in Beer

(reference: ASBC methods of Analysis, 8th Edition, 1992)

Method

1. Transfer 10.0 mL beer to a 50 mL centrifuge tube.
2. Add 50 uL octyl alcohol, 20 mL isooctane (HPLC grade) and 1 mL 3M HCl.
3. Shake vigorously for 15 minutes.
4. Centrifuge to separate the phases.
5. Read organic phase at 275 nm (1 cm cell) vs blank (20 mL isooctane, 50 uL octyl alcohol).

Notes:

- isooctane should have an Abs at 275 nm <0.005.
- readings should be done ASAP due to decomposition by UV light

Calculations:

BU= Abs at 275 nm*50

Example:

Abs =0.622
0.622*50= 31.1 BU

Alpha and Beta Acids in Hops

(reference: ASBC MoA. 8th edition, 1992)

Method

1. Place 5.000 +/- .001 gr pulverized hops in an extraction bottle and add 100 mL toluene.
2. Shake for 30 min with vigorous agitation.
3. Let stand until clear or centrifuge (preferred).
4. Dilution A: Dilute 5.0 ml of this extract to 100 mL with methanol.
5. Dilution B: Dilute an aliquot of the dilution A with alkaline methanol (0.2 mL 6M NaOH per 100 mL MeOH) so that the Abs at 325 and 355 falls within the most accurate range of the instrument.
6. Immediately read dilution B (1 cm) at 275, 325 and 355 vs a toluene blank that was prepared and diluted in EXACTLY the same manner.

Notes:

- Hexane may be substituted for toluene

Calculations:

Dilution factor, d= (volume dil A x volume dil B)/ (500 x aliq extract A x aliq dil A)

% alpha acids= d x (-51.56 A355+ 73.79 A325-19.07 A275)

% beta acids= d x (55.57 A355-47.59 A325 + 5.10 A275)

Example:

1. 5 gr hops extracted with 10 mL toluene
2. 5 mL clear extract diluted to 100 mL with methanol=Dilution A
3. 3 mL Dilution A diluted to 50 mL with alkaline methanol
4. Absorbances
    - A355=0.615
    - A325= 0.596
    - A275=0.132

d = (100 x 50) / (500 x 5 x 3) = 0.667

alpha = 0.667 x [-(51.56 x 0.615) + (73.79 x 0.596) - (19.07 x 0.132) = 6.5

beta = 0.667 x [(55.57 x 0.615) - (47.59 x 0.596) + (5.10 x 0.132) = 4.3

Alpha and Beta Acids in Hops by HPLC

(reference: ASBC MoA. 8th edition, 1992)

Method

1. Add 20 mL MeOH to 10.0 gr finely ground hops.
2. Add 100 mL diethyl ether.
3. Stopper and shake for 30 min.
4. Add 40 mL 0.1M HCl.
5. Stopper and shake for 10 min.
6. Allow to stand for 10 min to separate the phases.
7. Pipette 5.0 mL of the supernate to a 50 mL volumetric flask.
8. Bring to volume (50 mL) with methanol.
9. Filter before injecting (sample is stable 24 hours).
10. Calculate based on a known calibration standard as follows.

Notes:

- detector: capable of 314 nm
- column: C18 (they recommend 250x4mm, 5 um ODS, RP18, Nucleosil-5)
- Mobile phase: MeOH: H2O: HPO4 (85%) /85:17:0.25 (v/v)
- Conditions: isocratic
- Flow: 0.8 mL/min
- Temp: ambiant
- Sample: 10 uL
- Typical Retention times:
    - cohumulone 16 min
    - n- + ad-humulone 19 min
    - colupulone 27 min
    - n- + adlupulone 34 min

Calculations:

Response Factor, RF= [mass of calib extr (gr) x conc of component in calib extr (%)] / area.

Component %= (2 x average sample peak area of component x RF) / mass of sample