Ethanol breath analysis in seed testing

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New Developments and Technologies in Seed Testing

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Ethanol as marker for seed quality

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The volatile exudates from germinating pea seeds of different viability and vigor. Can. J. Bot. 63: 1035

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Stand Establishment/Seed

Eds. Liptay, Vavrina, Welbaum

Acta Hort. 504, ISHS 1999

ETHANOL PRODUCTION BY HYDRATED SEEDS: A HIGH RESOLUTION

INDEX OF SEED QUALITY

Wageningen Seed Centre

For quality of life
Ethanol measurement

Method Wayne T. Buckley (Agriculture and Agri-Food Canada, Brandon)

- Seeds + water in a vial
- Incubation
- Ethanol measured in the headspace. Modified breath analyzer (Dräger Alcotest 6810)

- Simple
- Fast
- Cheap
Seed vigour canola

Wayne T. Buckley, Brandon Research Centre, Canada
Seed vigour canola

Wayne T. Buckley, Brandon Research Centre, Canada
Mitochondrial damage

- In non-viable embryo’s of rye, rice and maize, the internal membrane structure of the mitochondria is distorted
Mitochondrial damage

Membrane damage due to aging (oxidation)

Electron micrographs of pea mitochondria

Control 12 hr imb.  Control 22h imb.  CD 22 hr imb.

A. Benamar et al. 2003
Aerobic respiration

\[ C_6H_{12}O_6 \rightarrow C_3H_4O_3 \]

glucose          pyruvate

\[ CO_2 + H_2O \]

Citric acid cycle in the mitochondria
aerobic respiration

\[ \text{C}_6\text{H}_{12}\text{O}_6 \rightarrow \text{C}_3\text{H}_4\text{O}_3 \]

Glucose \rightarrow Pyruvate

\[ \text{CO}_2 + \text{H}_2\text{O} \]

Citric acid cycle

in the mitochondria

\[ \text{O}_2 \rightarrow \text{O}^- \]

Acetaldehyde \rightarrow Ethanol

\[ \text{C}_6\text{H}_{12}\text{O}_6 \rightarrow \text{C}_3\text{H}_4\text{O}_3 \]

Glucose \rightarrow Pyruvate
Effect seed maturity on ethanol production

Seed (white cabbage) sorted based on chlorophyll content
Fraction 1: High chlorophyll, low maturity, low vigour.
Fraction 6: Low chlorophyll, High maturity, high vigour

Plant material: *Brassica oleracea* cv Bartollo F1, seeds. Ethanol Assay: 0.5 g, 30% MC
Effect seed maturity on ethanol production

Seed (white cabbage) sorted based on chlorophyll content
Fraction 1: High chlorophyll, low maturity, low vigour.
Fraction 6: Low chlorophyll, High maturity, high vigour

Plant material: Brassica oleracea cv Bartolo F1, seeds. Ethanol Assay: 0.5 g, 30% MC
Effect seed ageing on ethanol production

Ethanol production at 20°C reflects vigour decrease due to aging

At 40°C ethanol production becomes stronger with aging effect on germination

Which is even stronger with longer incubation time

Plant material: *Brassica oleracea*
Relation ethanol production and quality

- Asparagus
- Arabidopsis
- Barley
- Cabbage
- Canola
- Carrot
- Chinese cabbage
- Lettuce
- Onion
- Rice
- Sugarbeet
- Tomato
- ...

N.B. Within species genetic variation. Sometimes also variation between seed lots in response. But assay works very good for comparing effect of treatments with a single lot.
Effect seed ageing on ethanol production

No effect aging on ethanol production or germination

Plant material: *Brassica rapa*
Ethanol metabolism

$C_3H_4O_3$ (pyruvate) $\xrightarrow{PDC}$ $C_2H_4O$ (acetaldehyde) $\xrightarrow{ADH}$ $C_2H_6O$ (ethanol)

$CO_2$ $\xrightarrow{PDC}$ $C_2H_4O$ $\xrightarrow{ADH}$ $C_2H_6O$ (ethanol)

$NAD^+$ $\xrightarrow{ADH}$ $NADH$ $\xrightarrow{ALDH}$ $NAD^+$

$NADH$ $\xrightarrow{ALDH}$ $NAD^+$

$CH_3COOH$ (acetic acid)

$NAD^+$ $\xrightarrow{ALDH}$ $NADH$

ADP $\rightarrow$ ATP

mitochondrion
Natural aging $\rightarrow$ ethanol degradation (aging)

Chinese Cabbage

- No effect aging on ethanol production or germination
- Clear effect aging on ethanol degradation

Plant material: *Brassica rapa*
Conclusions ethanol assay

- Clear correlation seed quality and ethanol production
- Different types of reduced quality correlate with increased ethanol production
  - Less maturity
  - Heat treatment
  - Natural aging
  - Conditional deterioration
- Capacity to degrade ethanol can be an additional assay for seed quality
Ethanol breath analysis in seed testing
Contact

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