Introduction

Nondestructive Infra Red Light High-throughput Biochemical Analysis of Single Seed Canola

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Introduction

Omega-9 Canola oil is the ideal product for the rapidly growing new segment of healthy, naturally-stable vegetable oils

Omega-9 Oils™
Healthier Oils. Healthier Business.™

*Trademark of Dow AgroSciences LLC

Introduction

Omega-9 Canola Oil Products Match Consumer Need

- Food industry seeking oxidative stability have used partially hydrogenated vegetable oils
  - Eight billion pounds of partially hydrogenated oils used in U.S. (overall market value approximately $2.8B)
- FDA announces mandatory labeling of trans fats (Jan. 06')
- Omega-9* canola increases content of "good" fats and reduces "bad" fats, while providing critical functionality
- Decreased risk of coronary heart disease associated with "bad fats"

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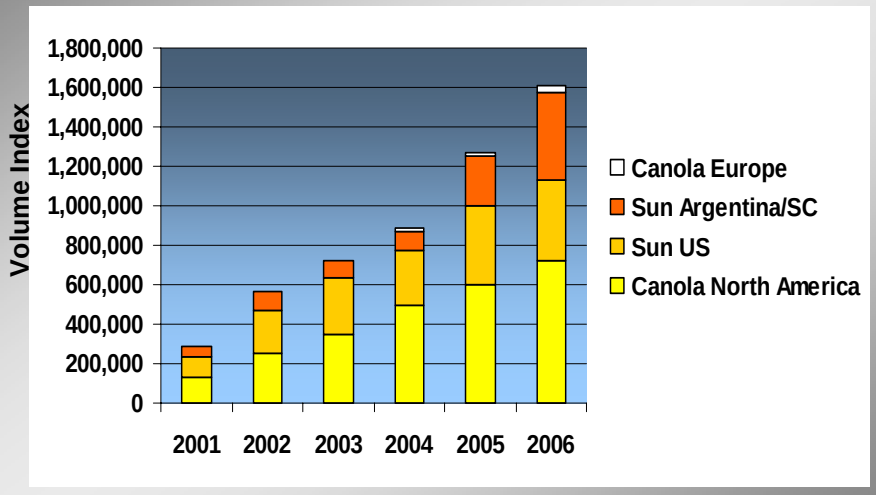
Growth of Dow AgroSciences Omega-9 Oils Program



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Introduction

Summary

- **Automation: hardware and software**
- **Chemometrics: model prediction**
- **Improvements: new instrumentation**
- **Application: large population screening**

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Automation

Generation of canola single seed NIR spectra

OPUS software driving data acquisition

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Automation

Generation of canola single seed NIR spectra

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Automation

Efficiency achieved in single seed

- **Per Tecan RSP150 in 24 hours period**
  - **throughput increased by 35% (1800 to 2250 seed from envelopes)**
  - **throughput increased by 50% (2880 seeds from plates)**
  - **missing seed decreased using new vacuum chip (1% from plate to plate to 2% from envelope)**

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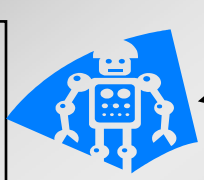
Automation

Software/Hardware Interactions

- Gemini controls robot
- VB program triggered by Gemini
- VB program triggers Opus
- Opus scans spectrum
- InStep™ triggered by VB
- Gemini resumes when VB complete



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Chemometrics

Calibration Model Development

- Plot the data
  - Identify obvious problem data
- Construct model
  - Select a preprocessing method
  - Select samples (outlier detection)
  - Select variables (wavelength selection)
- Validate model
  - Examine calibration diagnostics
  - Assess performance
    - cross validation often used
    - separate test set prediction

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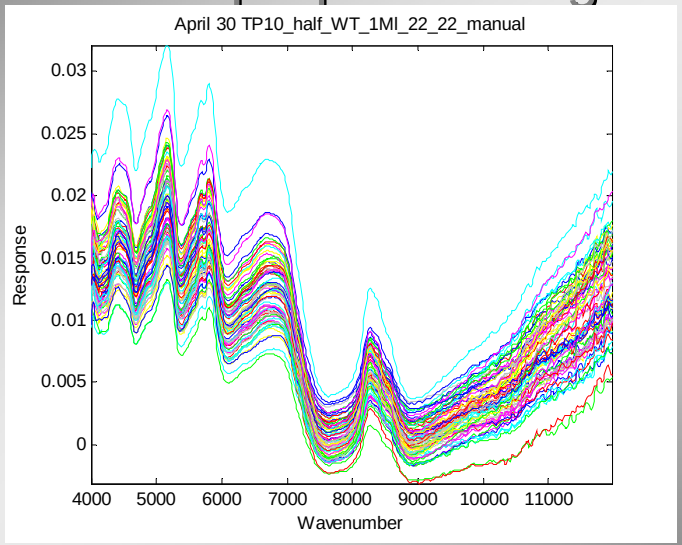
Chemometrics

Select preprocessing



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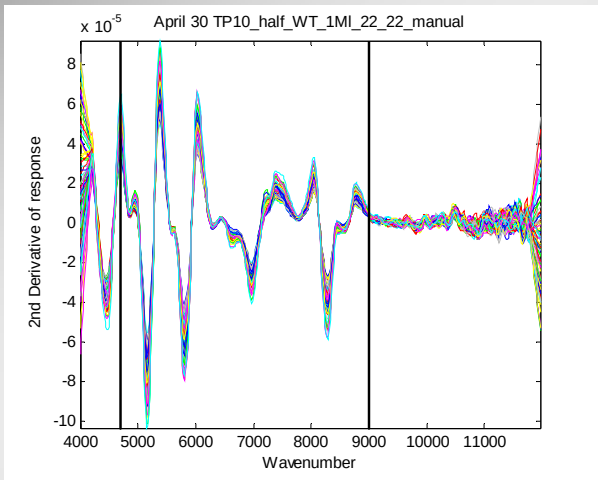
Chemometrics

2nd derivative of NIR data

25 point window, 3rd order polynomial



Chemometrics

Validation

Cross validation

	Sample # 1	Sample # 2	Sample # 3	Sample # 4	Sample # 5	Sample # 6	Sample # 7	Sample # 8
Cycle #1	V	C	C	C	C	C	C	C
Cycle #2	C	V	C	C	C	C	C	C
Cycle #3	C	C	V	C	C	C	C	C
Cycle #4	C	C	C	V	C	C	C	C
Cycle #5	C	C	C	C	V	C	C	C
Cycle #6	C	C	C	C	C	V	C	C
Cycle #7	C	C	C	C	C	C	V	C
Cycle #8	C	C	C	C	C	C	C	V

- This is a graphic example of a LOO-CV experiment.
- Eight samples lead to 8 calibration and prediction cycles
- Each sample left out (put in validation) once and only once.
- Should be possible to execute in any reasonable software package.

Summary

- **Automation: hardware and software**
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- **Improvements: new instrumentation**
- **Application: large population screening**

Improvements

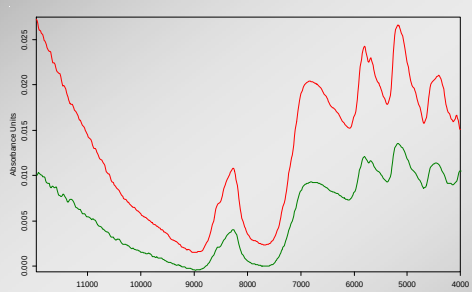
Acquisition of new FTNIR: MPA

- Two fiber optic probe
- Multi purpose analyzer (transmission and reflection)
- New version of OPUS
- Same rugged interferometer

Improvements

System modification

- New integrated sphere (8% intensity gain)
- High intensity source (2.5 X signal amplitude)



- Dry N_2 purged instrument to eliminate residual moisture increase reliability and stability
- New version of OPUS (firmware update)

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Applications

Generation of canola single seed reference methods

- Quantitative and qualitative analysis require
  - accurate weighing (0.01 mg)
  - exhaustive extraction (multiple)
  - automated pipetting (Hamilton)
- Analytical instrument
  - GC/FID
  - HPLC/Fluorescence/ELSD/UV
- Sensitivity and reproducibility
  - Medium throughput (96 well plates)
  - Derivatization to increase sensitivity

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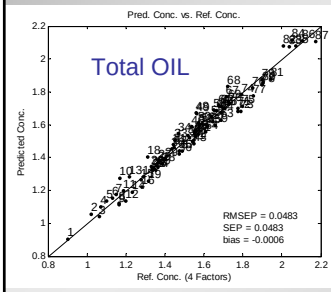
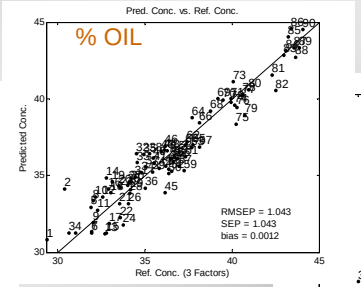
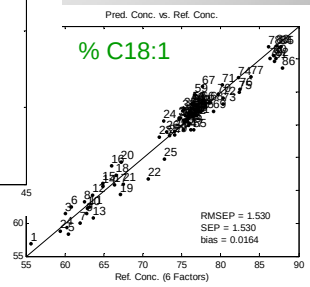
Applications

Prediction Performance for oil

Factor	RMSEP	SEP	r2	Range/RMSEP	SD/RMSEP
4*	0.0483	0.0483	0.9679	26.3	5.61
3*	1.0429	1.0429	0.9138	14.02	3.42
6*	1.53	1.5299	0.9588	21.09	4.95







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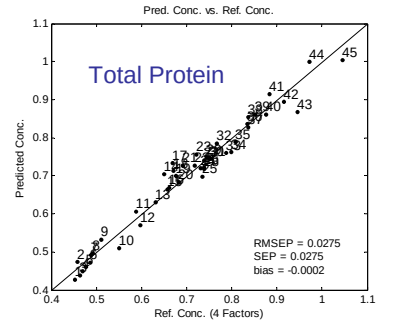
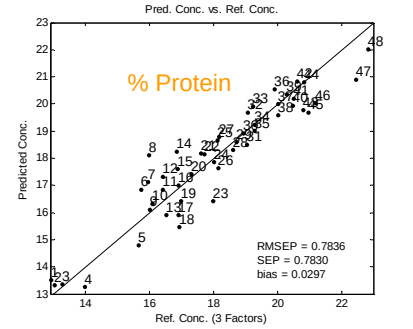
Applications

Prediction Performance for Protein

Factor	RMSEP	SEP	r2	Range/RMSEP	SD/RMSEP
4*	0.0275	0.028	0.966	21.6	5.49
3*	0.7836	0.783	0.884	12.62	2.96





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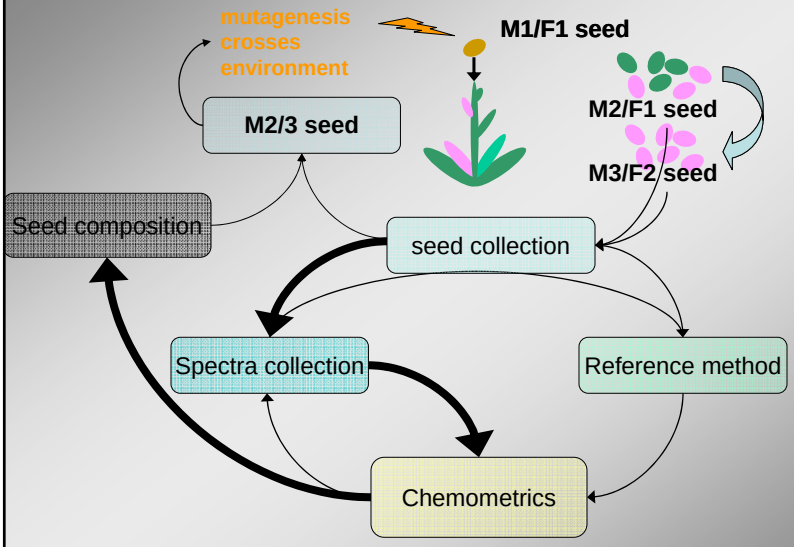
Applications

Single seed screening process



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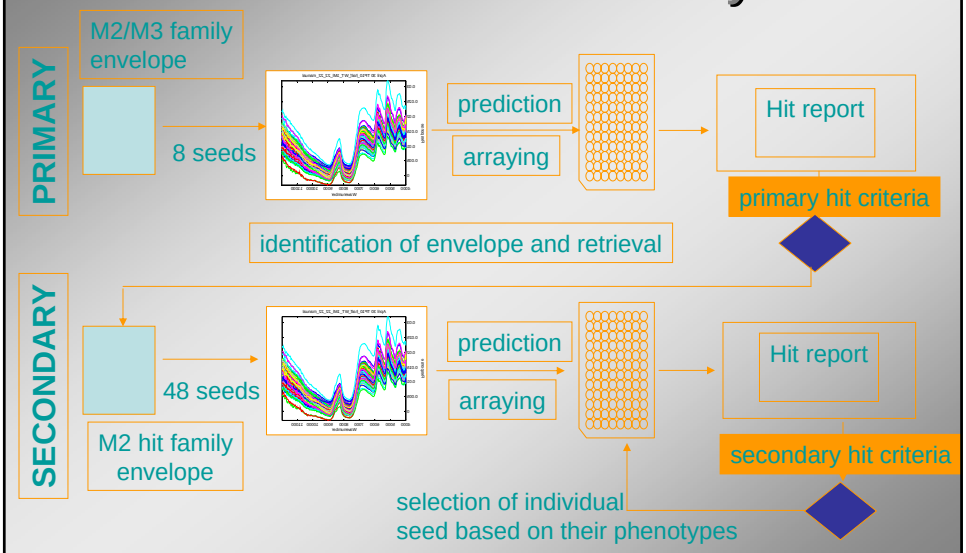
Applications

Selection scheme for hits by FTNIR



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Thank you for your attention

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