



**Agricultural Innovation Program
Research Project Final Report**
Contribution Agreement - Vote 10 Funding

Project Title:	Development of a Designer Soybean Testing Methodology Activity 3: Essential Phytochemical Compound Identification and Compound/Constituent Validation
Start Date (yyyy-mm-dd):	2012-04-01
Expected End Date (yyyy-mm-dd):	2013-03-31
Actual End Date (yyyy-mm-dd):	2013-03-31
Principal Investigator (PI):	Sevita International – Jim McCullagh
Short Executive Summary of report:	
<p>ECODA's major soybean industry partner, Sevita International, exports a wide range of soybean varieties to Japan for use by Japanese processors to make soymilk and tofu. Each Japanese customer has a different formulation, process and style of product and, therefore, different soybean varieties work better for some customers than others.</p> <p>To understand the relationship of a soybean variety and the end product, the Guelph Food Technology Centre (GFTC) performed soymilk and tofu sensory evaluations. Japanese processing clients of Sevita International visited the GFTC to consult on sensory evaluation techniques and provide their feedback on varieties being tested.</p> <p>The GFTC panelists participated in the evaluation of Japanese imported soymilks (two varieties) and GFTC-produced soymilk (five varieties using beans provided by Sevita International). Sensory evaluation results of the Japanese imported soymilks indicated a significant difference in the attributes of (a) raw green, (b) cooked bean, (c) sweet and (d) bitter between the samples, however, no differences in (e) astringency were observed. (The lexicon for these attributes was developed in Activity 1). Based on the colour measurements, a perceptual difference may exist between the two imported Japanese soymilk samples. Sensory evaluation of the GFTC-produced soymilks indicated significant differences among the five varieties for sweetness. OAC Champion was significantly less sweet than Kinusayaka and S03W4. No differences in intensity were observed for the attributes of raw green, cooked bean, bitter and astringent. Based on the colour measurements, a perceptual difference may exist between the soymilk samples with the exception of the OAC Champion/S03W4 and the Kinusayaka/DH530 samples.</p> <p>The GFTC panelists also participated in the evaluation of six varieties of tofu produced at GFTC using beans provided by Sevita International. Sensory evaluation of the GFTC-produced tofu indicated a significant difference between OAC Champion and DH410SCN for the attribute of beaniness. Based on the colour measurements, a perceptual difference may exist between the tofu samples with the exception of Stargazer-DH420, DH410SCN-S03W4 and DH410SCN-DH420 samples. Although not significant, the different tofu samples exhibited three rough groups of firmness. Samples DH530 and DH420 were consistently the two firmest samples, followed by the mid-range firmness of samples DH410SCN and S03W4. Samples OAC Champion and Stargazer were consistently less firm than any of the other samples that were tested.</p> <p>The responses from the trained panelists were quantified and this data, along with samples of each end product, were sent to the University of Ottawa for analysis in their ultra-high performance liquid chromatography with time-of-flight mass spectrometry (UPLC MS QTOF) analytical facility. Putative identification of specific taste and odour components was achieved by performing discriminant analysis based on human panel data to differentiate variety. A number of biomarkers were identified by discriminate analysis in raw beans and processed tofu or soymilk. Biomarkers that differentiate varieties with significant human panel differences were found. In conclusion, the project has met the objectives of identifying the soy metabolome and employing statistical methods to identify phytochemical biomarkers for variety comparisons and taste preference. The project was able to successfully match preferred commercial varieties with unique Sevita International varieties using metabolomic data.</p> <p>To ensure that the variety recommendations obtained from the University of Ottawa would meet tofu production requirements, Sevita International sent samples of the recommended varieties to a Japanese tofu testing laboratory for analysis.</p>	



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A. Research Progress and Accomplishments (to date in relation to expected milestones and deliverables / outputs)

- Include brief summary of:
 - Introduction, literature review, objectives, milestones and deliverables / outputs.
 - Approach / methodology (summary by objectives).
- Include results and discussion (overview by objectives and milestones), next steps and references.

Introduction

ECODA's major soybean industry partner, Sevita International, exports a wide range of soybean varieties to Japan for use by Japanese processors to make soymilk and tofu. Each Japanese customer has a different formulation, process and style of product and therefore different bean varieties work better for some customers than others.

In Activity 1, the GFTC used a combination of literature searches, individual expertise, professional consultation and feedback provided by Sevita International and the Japanese collaborators to develop the a standardized process for making both soymilk and tofu as well as the lexicon required for evaluating the end products using a trained sensory panel.

In Activity 2, the University of Ottawa performed non-targeted compositional testing on soybean varieties that were either preferred or not preferred by soy-food manufacturers as well as soyfood products made with known soybean varieties.

Activity 3 builds on these first two activities by performing soymilk and tofu sensory evaluations at the GFTC to understand the relationship that a soybean variety has on an end product. The responses from the trained panelists were quantified and this data was sent to the University of Ottawa for analysis. Discriminant analysis was used to identify specific taste and odour components. These components, along with the results from the Japanese tofu test results, were then used to help facilitate the recommendation of new replacement varieties for Sevita International's customers. Japanese collaborators visited the GFTC during the sensory evaluations to consult on sensory evaluation techniques and provide their feedback on varieties being tested.

Objectives

- Link chemical compounds in soybeans to sensory evaluations by conducting an analysis of test results from raw soybeans and finished soy-food products.
- Validation of whether compounds found to affect sensory evaluations will impart the same effect when found in other varieties.
- Supply new varieties that contain the essential compounds for testing by the end user and sensory evaluation.

Deliverables

- A summary of findings.

Methodology - Soymilk

Two varieties of soymilk from Japan were provided by Sevita International to the GFTC for the purpose of sensory evaluation. The imported soymilk varieties were:

- Kinusayaka (Blue Tetra Pak)
- DH530 (White Tetra Pak)

Five varieties of soybeans were provided by Sevita International to the GFTC for the purpose of producing and evaluating soymilk. The soybean varieties were:

- DH530
- DH618
- OAC Champion
- S03W4
- Kinusayaka

Sensory Panel Selection

Potential candidates were identified for this activity through email responses to a general prescreening questionnaire (Figure 1).



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Figure 1: General questionnaire used for screening potential sensory panel candidates



GFTC will be conducting trained sensory panels for soy based food products from September, 2012 to January, 2013. We are looking for participants for our panel. If you are interested in participating and meet the following qualifications please respond to Karen McPhee, kmcphee@gftc.ca:

1. Interest in full participation in the sensory training and evaluations
2. Availability to participate in 80% or more of all phases of the panels' work
3. No food allergies to soy (if you have other allergies please indicate in your reply)
4. Have general good health and no illnesses related to sensory properties being measured such as;
 - a. Diabetes,
 - b. Hypoglycaemia,
 - c. Hypertension,
 - d. Dentures,
 - e. Chronic colds

Details of Panel

Start Day: September 10 2012

Day: Monday, Wednesday, Friday

Time: 11:00am -12:00pm (1 hour)

End Date: January 30, 2012

Remuneration: \$15/session

Your participation would be appreciated!

Upon meeting the required criteria from the general questionnaire, potential panelists were asked to participate in a triangle difference test. The triangle difference test consisted of two evaluations of soymilk, panel A and panel B. These two evaluations were conducted simultaneously. Refer to Table 1 for samples presented to panelists.



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Table 1. Soymilk samples presented to potential panelists for sensory panel selection.

	Panel A	Panel B
Control sample	Soymilk (DH530, White Tetra Pak)	Soymilk (DH530, White Tetra Pak)
Test sample	99.25% Soymilk (DH530, White Tetra Pak) plus 0.75% sugar	99.5% Soymilk (DH530, White Tetra Pak) plus 0.5% soy bean solution

For panel A and B, respectively, half of the panelists received three samples consisting of two control samples and one test sample and the other half of panelists received three samples consisting of one control sample and two test samples. All samples were labeled with a three digit blinding code to prevent testing bias from occurring. Panelists were asked to identify the odd sample in panel A and panel B. See Figure 2 for the triangle difference ballot used in the triangle difference test.

Figure 2: Triangle difference ballot for soymilk

Name _____ Date _____

Product: Soymilk

Two of these samples are identical, and one may be different.
Please evaluate for taste differences.

- Evaluate the samples in the order presented and identify the odd sample. Be sure to identify one sample as different.

3 Sample Codes: _____, _____, _____

Odd sample Code: _____
- Indicate the degree of difference between the duplicate samples and the odd sample.

Slight _____ Moderate _____ Much _____ Extreme _____
- Comments:

Please describe the characteristics of the duplicate sample:

Please describe the characteristics of the odd sample:

Panelists to be trained for sensory evaluation of soymilk were selected based on the completion of triangle difference tests, panel A and panel B, respectively. Based on the results of the triangle difference tests, a total of 13 panelists were selected to be part of the trained sensory panel for the evaluation of soymilk.

- Brenda Simmons 13-5-30 7:33 PM
- Comment [1]:** On Page 2, doesn't it say white tetra pak?
- Brenda Simmons 13-5-30 7:33 PM
- Comment [2]:** Same question



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Soy milk Sensory Panel Training & Sensory Evaluation

Selected panelists were trained on pre-determined attributes (Table 2). For training purposes, panelists were presented with solutions representing each attribute at three intensity levels: low, medium and high.

Table 2. Attribute and attribute definitions for soymilk evaluation.

Attribute	Definition
Raw Green	aromatic characteristics of freshly cut green beans
Cooked Bean	aromatic characteristics of cooked beans or soy beans
Sweet	taste on the tongue stimulated by sugars
Bitter	taste on tongue stimulated by solutions of caffeine, quinine and certain other alkaloids
Astringent	the chemical feeling factor on the tongue or other skin surfaces of the oral cavity described as puckering/drying and associated with tannins or alum

Table 3 below outlines each attribute and varying degrees of intensity. The solutions in Table 3 were also used as the reference samples during soymilk sensory evaluation panels. This step of the training process helps to standardize panelist responses to various attributes.

Table 3. Solutions used for training purposes and as reference samples.

	Raw Green	Cooked Bean	Sweet	Bitter	Astringent
Low	Water	Water	Water	Water	0.025% Alum Solution
Mid	50% Raw Green Stock Solution	25% Cooked Bean Stock Solution	0.75% Sugar Solution	0.015% Bitter Stock Solution	0.0625% Alum Solution
High	100% Raw Green Stock Solution	50% Cooked Bean Stock Solution	1.5% Sugar Solution	0.04% Bitter Stock Solution	0.1% Alum Solution

Solution Preparation & Ingredient Specifications

Preparation of soybean solution for triangle difference test

1. Soak dried soybeans (variety DH530) for 24 hours.
2. Rinse and strain soybeans.
3. Grind soybeans (300g) with spring water (100g) using a Cuisinart hand blender (approximately 2 minutes).
4. Filter solution using cheesecloth to filter out soybean particulates.
5. Use solution to spike samples of soymilk for the panel B triangle difference test (at 0.5%).

Preparation of raw green bean solution for sensory training and soymilk evaluation

1. Wash and drain fresh green beans (variety DH530).
2. Cut green beans into 0.5cm pieces.
3. Place 155g of cut green beans into a glass jar and add 465g of spring water.
4. Seal jar and refrigerate for 24 hours.
5. Filter water from green beans using a cheese cloth.
6. Use filtered water as raw green stock solution.



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Preparation of cooked bean solution for sensory training and soymilk evaluation

1. Rinse dried soybeans (variety DH530).
2. Soak soybeans in water overnight (approximately 24 hours).
3. Rinse and drain soaked soybeans.
4. In large pot, combine 450g soaked beans and 2.5kg spring water.
5. Bring soybeans and water to a boil and boil for one hour.
6. Filter water from soybeans using a cheese cloth.
7. Use filtered water as cooked bean stock solution.

Table 4: Preparation of sweet solution for sensory training and soymilk evaluation

Solution	Low	Medium	High
Sweet	100% Spring water	0.75% Sugar + 99.25% Spring water	1.5% Sugar + 98.5% Spring water
Bitter*	100% Spring water	0.015% Bitter stock solution* + 99.99% Spring water	0.04% Bitter stock solution* + 99.96% Spring water
Astringent	0.025% Alum + 99.98% Spring water	0.0625% Alum + 99.94% Spring water	0.1% Alum + 99.90% Spring water

Preparation of 0.1% bitter stock solution

1. Bring to a boil 99.9% spring water.
2. Add 0.1% caffeine and stir.
3. Remove bitter stock solution from heat and cool to room temperature.

Training Sessions

A panel of 13 individuals was trained using attributes generated during the lexicon development (Refer to Activity 1 - Lexicon Development). Training was undertaken to define these attributes as they related to the soymilk samples. Definitions of each attribute are shown in Table 2.

During training sessions, panelists were presented with four varieties of soymilk sub-sampled from soymilk products produced at the GFTC and using soybean varieties provided by Sevita International. Soymilk samples were provided in 1oz covered Dixie sample cups and labelled with a three digit blinding code to prevent testing bias from occurring. Samples were provided in a non-randomized order for discussion purposes. Each training session focussed on the discussion of one to two attributes. Training sessions involved open discussions on the definition of each attribute, where the attribute is detected in the mouth, when the attribute is detected and when the specific attribute is at its peak (i.e. the strongest).

For training purposes and soymilk evaluation, panelists were asked to use the following tasting method/protocol:

- Take a sip of soymilk
- Hold soymilk in mouth for three seconds
- Exhale (out of nose)
- Swallow soymilk
- Evaluate sample for specified attribute (Note: panelists were asked to wait 30 seconds prior to making an evaluation for bitter and astringency attributes only, as these attributes build with time)

For all attributes, excluding astringency, panelists were asked to cleanse their pallet with crackers and water in between samples. For the astringency attribute, panelists were asked to cleanse their pallet with unsweetened apple sauce (Brand: President’s Choice Unsweetened Applesauce) and water to minimize the buildup of mouth dryness.


At the end of each training session, panelists were provided with the opportunity to practice evaluating soymilks for intensity of each attribute. Panelists were presented with four GFTC produced soymilk varieties labelled with a three digit blinding code to prevent testing bias from occurring. Approximately 28ml of soymilk per sample were provided in 1oz covered Dixie sample cups. Using a 10cm line scale, panelists were asked to evaluate each sample using the ballot (as seen in Figure 3) provided and to evaluate the products based on the reference samples (Table 3) provided for each



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attribute.

Figure 3: Training and sensory evaluation ballot of soymilk



Product: Soy Milk Product ID: _____

Judge # _____

Name: _____ Date: _____

Please evaluate the attributes below for soy milk. Make a vertical line on the horizontal line to indicate your rating of the attribute.

Raw Green:

Low Raw Green Mid Raw Green High Raw Green

Cooked Bean:

Low Cooked Bean Mid Cooked Bean High Cooked Bean

Sweet:

Low Sweetness Mid Sweetness High Sweetness

Bitter:

Low bitter Mid Bitter High Bitter

Astringent:

Low Astringency Mid Astringency High Astringency

Sensory Panel Evaluation

After training was completed, panelists participated in the evaluation of:

- 1.) Japanese imported soymilks, two varieties
- 2.) GFTC-produced soymilk, five varieties, using soybeans varieties provided by Sevita International

During evaluation sessions, soymilk samples were presented in partitioned booths for evaluation. All testing was



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completed using a 10cm line scale. Three replications of testing of each sample were conducted. During each replicate of testing, 28ml of each sample was served in a randomized order to the panelists. All samples were labelled with a three digit blinding code to prevent testing bias from occurring. Samples were evaluated at 15°C. For each session, panelists were asked to evaluate soymilk samples for intensity of each attribute using the ballots provided (one ballot per product) and evaluate the products using the reference samples provided for each attribute. Refer to Figure 3 for the ballot used during sensory evaluation. During sensory evaluation, panelists used the tasting method/protocol described above.

Statistical Design and Analysis

Upon completion of the sensory evaluation of soymilk, each marked line across the 10cm line scale was measured and data (to one decimal place) was recorded. To determine differences in intensity of each attribute between or among the samples, an analysis of variance (ANOVA) test was conducted. Where differences in mean scores were observed, a Tukey's Honestly Significant Difference (HSD) test was conducted to determine where the differences were. Significance is reported at 95% confidence.

Colour Analysis

A Minolta CR-300 Colorimeter was used to objectively measure the lightness (i.e. the L value expressed as 0 for black to 100 for white), degree of red to green (i.e. the a value with increasingly positive values approaching red and increasingly negative values approaching green) and the degree of yellow to blue (i.e. the b value with increasingly positive values approaching yellow and increasingly negative values approaching blue). For each day of sensory evaluation, a colour measurement was taken. Soymilk was poured into glass test tubes and the measuring head of the colorimeter was positioned on three different locations of the test tube. The reported result is the average of the readings from each day of testing. For more information on Minolta colour measurement please refer to konicaminolta.com.

Methodology - Tofu

Six varieties of soybeans were provided by Sevita International to the GFTC for the purpose of producing and evaluating tofu. The soybean varieties were:

- OAC Champion
- S03W4
- DH530
- DH410SCN
- DH420
- Stargazer

Sensory Panel Selection

Potential candidates were initially screened by completing a general questionnaire to determine if they would be qualified panelists. Refer to Figure 1.

Upon meeting the required criteria from the general questionnaire, potential panelists were asked to participate in a triangle difference test. The triangle difference test consisted of two evaluations of soymilk, panel A and panel B. These two evaluations were conducted simultaneously. Refer to Table 1 for samples presented to panelists.

For panel A and B, respectively, half of the panelists received three samples consisting of two control samples and one test sample and the other half of panelists received three samples consisting of one control sample and two test samples. All samples were labeled with a three digit blinding code to prevent testing bias from occurring. The panelists were asked to identify the odd sample in panel A and panel B. See Figure 4 for the triangle difference ballot used in the triangle difference test.

Panelists to be trained for sensory evaluation of tofu were selected based on the completion of soymilk triangle difference tests, panel A and panel B, respectively. Based on the results of the triangle difference tests, a total of 13 panelists were selected to be part of the trained sensory panel for the evaluation of tofu (Note: Due to scheduling conflicts, only 12 panelists participated in the tofu evaluation).



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Figure 4: Triangle difference ballot for tofu

Name _____ Date _____

Product: Tofu

Two of these samples are identical, and one may be different.
Please evaluate for taste differences.

1. Evaluate the samples in the order presented and identify the odd sample. Be sure to identify one sample as different.

3 Sample Codes: _____, _____, _____

Odd sample Code: _____

2. Indicate the degree of difference between the duplicate samples and the odd sample.

Slight _____ Moderate _____ Much _____ Extreme _____

3. Comments:

Please describe the characteristics of the duplicate sample:

Please describe the characteristics of the odd sample:

Tofu Sensory Panel Training & Sensory Evaluation

Solution Preparation

Selected panelists were trained on pre-determined attributes (Table 5). For training purposes, panelists were presented with solutions representing each attribute at three intensity levels: low, medium and high.

Table 5. Attribute and attribute definitions for tofu evaluation

Attribute	Definition
Bean	aromatic characteristics beans or soy beans
Sweet	taste on the tongue stimulated by sugars
Bitter	taste on tongue stimulated by solutions of caffeine, quinine and certain other alkaloids
Astringent	the chemical feeling factor on the tongue or other skin surfaces of the oral cavity



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[] described as puckering/drying and associated with tannins or alum

Table 6 below outlines each attribute and varying degrees of intensity. The solutions in Table 3 were also used as the reference samples during tofu sensory evaluation panels.

Table 6. Solutions Used for Training Purposes and Sensory Evaluation of Tofu

	Bean	Sweet	Bitter	Astringent
Low	Spring Water	Spring Water	Spring Water	Spring Water
Mid	12.5% Raw Green Stock Solution 37.5% Cooked Bean Stock Solution 50 % Spring Water	0.75% Sugar Solution	0.015% Bitter Stock Solution	0.04% Alum Solution
High	25% Raw Green Stock Solution 75% Cooked Bean Stock Solution	1.5% Sugar Solution	0.04% Bitter Stock Solution	0.08% Alum Solution

Solution Preparation & Ingredient Specifications

Preparation of soybean solution for triangle difference test

1. Soak dried soybeans (variety DH530) for 24 hours.
2. Rinse and strain soybeans.
3. Grind soybeans (300g) with spring water (100g) using a Cuisinart hand blender (approximately 2 minutes).
4. Filter solution using cheesecloth to filter out soybean particulates.
5. Use solution to spike samples of tofu for the panel B triangle difference test (at 0.5%).

Preparation of raw green bean solution for sensory training and tofu evaluation

1. Wash and drain fresh green beans (variety DH530).
2. Cut green beans into 0.5cm pieces.
3. Place 155g of cut green beans into a glass jar and add 465g of spring water.
4. Seal jar and refrigerate for 24 hours.
5. Filter water from green beans using a cheese cloth.
6. Use filtered water as raw green stock solution.

Preparation of cooked bean solution for sensory training and tofu evaluation

1. Rinse dried soybeans (variety DH530).
2. Soak soybeans in water overnight (approximately 24 hours).
3. Rinse and drain soaked soy beans.
4. In large pot, combine 450g soaked beans and 2.5kg spring water.
5. Bring soybeans and water to a boil and boil for one hour.
6. Filter water from soy beans using a cheese cloth.
7. Use filtered water as cooked bean stock solution.

Table 7: Preparation of solutions for sensory training and tofu evaluation

Solution	Low	Medium	High
Sweet	100% Spring water	0.75% Sugar + 99.25% Spring water	1.5% Sugar + 98.5% Spring water
Bitter*	100% Spring water	0.015% Bitter stock solution* + 99.99% Spring water	0.04% Bitter stock solution* + 99.96% Spring water
Astringent	100% Spring water	0.04% Alum + 99.96% Spring water	0.08% Alum + 99.92% Spring water

Brenda Simmons 13-5-30 7:37 PM
Comment [3]: What is the asterisk for?



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Preparation of 0.1% bitter stock solution

1. Bring to a boil 99.9% spring water.
2. Add 0.1% caffeine and stir.
3. Remove bitter stock solution from heat and cool to room temperature.

Training Sessions

A panel of 12 individuals was trained using attributes generated during the lexicon development (refer to Activity 1 - Lexicon Development). Training was undertaken to define these attributes as they related to the tofu samples.

Definitions of each attribute are shown in Table 5.

During training sessions, panelists were presented with four varieties of tofu sub-sampled from tofu products produced at GFTC using bean varieties provided by Sevita International. Tofu samples were provided in 2oz covered Dixie sample cups and labelled with a three digit blinding code to prevent testing bias from occurring. Samples were provided in a non-randomized order for discussion purposes. Each training session focussed on the discussion of one to two attributes. Training sessions involved open discussions on the definition of each attribute, where the attribute is detected in the mouth, when the attribute is detected and when the specific attribute is at its peak (i.e. the strongest).

For training purposes and tofu evaluation, panelists were asked to use the following tasting method/protocol:

- Place one piece of tofu on the front of the tongue and push the tofu up towards the roof of the mouth
- Break the tofu three times with the tip of the tongue against the roof of the mouth
- Move tofu around in the mouth
- Breath out of nose
- Swallow

For all attributes, excluding astringency, panelists were asked to cleanse their palate with crackers and water in between samples. For the astringency attribute, panelists were asked to cleanse their palate with unsweetened apple sauce (Brand: President's Choice Unsweetened Applesauce) and water to minimize the buildup of mouth dryness.

For the evaluation of bitterness and astringency, panelists were instructed to wait 30 seconds after swallowing the tofu to make the selection on the ballot since these attributes may not be detected immediately after swallowing and tend to build with time.

At the end of each training session, panelists were provided with the opportunity to practice evaluating tofu samples for the intensity of each attribute. Panelists were presented with four GFTC produced tofu varieties labelled with a three digit blinding code to prevent testing bias from occurring. Six pieces (approximately 0.6cm³) of tofu were provided in 2oz covered Dixie sample cups. Using a 10cm line scale, panelists were asked to evaluate each sample using the ballot provided and to evaluate the products based on the reference samples (Table 3) provided for each attribute. See Figure 5 for the ballot used during training sessions.

Sensory Panel Evaluation

After training was completed, panelists participated in the evaluation of six varieties of GFTC-produced tofu using bean varieties provided by Sevita International.

During evaluation sessions, tofu samples were presented in partitioned booths for evaluation. All testing was completed using a 10cm line scale. Three replications of testing of each sample were conducted. During each replicate of testing, six pieces (approximately 0.6cm³) of each tofu variety were served in a randomized order to panelists. All samples were labelled with a three digit blinding code to prevent testing bias from occurring. Samples were evaluated at 10-15°C. For each session, panelists were asked to evaluate tofu samples for intensity of each attribute using the ballots provided (one ballot per product) and evaluate the products using the reference samples provided for each attribute.

Refer to Figure 5 for the ballot used during sensory evaluation. During sensory evaluation, panelists used the tasting method/protocol described above. Since the tofu evaluation consisted of six products, panelists were instructed to evaluate three tofu products, wait 10 minutes to rest the palate, and then continue with the evaluation of the remaining three tofu products.



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Figure 5: Training and sensory evaluation ballot of tofu



Product: Tofu

Product ID: _____

Judge # _____

Name: _____

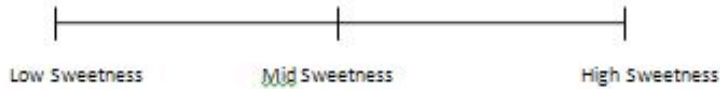
Date: _____

Please evaluate the attributes below for tofu. Make a vertical line on the horizontal line to indicate your rating of the attribute.

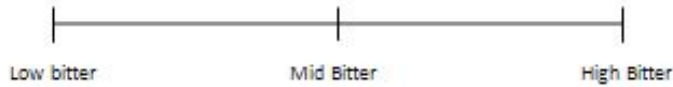
Bean:



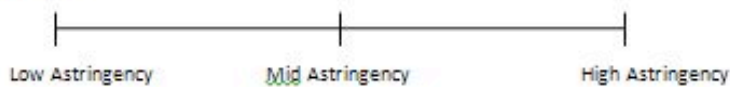
Sweet:



Bitter:



Astringent:





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Statistical Design and Analysis

Upon completion of the sensory evaluation of tofu, each marked line across the 10cm line scale was measured and data (to one decimal place) was recorded. To determine differences in the intensity of each attribute between or among the samples, an analysis of variance (ANOVA) test was conducted. Where differences in mean scores were observed, a Tukey's Honestly Significant Difference (HSD) test was conducted to determine where the differences were. Significance is reported at 95% confidence.

Colour Analysis

A Minolta CR-300 Colorimeter was used to objectively measure the lightness (i.e. the L value expressed as 0 for black to 100 for white), degree of red to green (i.e. the a value with increasingly positive values approaching red and increasingly negative values approaching green) and the degree of yellow to blue (i.e. the b value with increasingly positive values approaching yellow and increasingly negative values approaching blue). For each day of sensory evaluation, a colour measurement was taken. Tofu was sliced into approximately 8cm x 4cm x 0.5cm and the measuring head of the Colorimeter was positioned on three different locations of the tofu slice. The reported result is the average of the readings from each day of testing. For more information on Minolta colour measurement please refer to konicaminolta.com.

Texture Analysis

The texture of each tofu variety was measured using a TA.XT.Plus Texture Analyzer with a 10mm diameter probe and using a 15mm penetration depth. Two replicates from each of three blocks for each sample per test day for a total of six measurements per day for each sample. A total of 18 measurements per sample were taken over the duration of the study. All sample blocks were divided into six smaller test blocks of approximately 4cm x 4cm x 2.5cm. Test blocks were blotted for approximately five seconds to remove excess moisture. Test blocks were consistently placed on the testing platform such that the height of the sample was 4cm. All tests were performed on test blocks pulled directly from refrigerated storage. Test blocks not used for immediate texture analysis were placed back into refrigerated storage for later testing.

Results & Discussion - Soymilk

Sensory Evaluation of Japanese Imported Soymilk

Mean scores for the samples imported from Japan are shown in Table 8. Significant differences in raw green, cooked bean, sweet and bitter were observed between the samples. No differences in astringency were observed.



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Table 8. Mean scores and Tukey's HSD results for sensory properties of Japanese imported soymilks

Attributes		Samples	
		A	B
Raw Green	Mean	4.5a ^{1,2}	2.4b
	St Error	0.36	0.26
Cooked Bean	Mean	4.9a	7.1b
	St Error	0.29	0.32
Sweet	Mean	4.6a	3.3b
	St Error	0.28	0.29
Bitter	Mean	2.5a	3.5b
	St Error	0.32	0.40
Astringent	Mean	3.2a	3.2a
	St Error	0.34	0.35

¹ Means in a row with the same letter are not significantly different (P<0.05)
² n=38

Sample A – DH530 (Blue Tetra Pak)

Sample B – Kinusayaka (White Tetra Pak)

Colour Evaluation of Japanese Imported Soymilk

Table 9 below provides the average scores for colour evaluation, where the lightness (i.e., the **L** value expressed as 0 for black to 100 for white), the degree of red to green (i.e. the **a** value with increasingly positive values approaching red and increasingly negative values approaching green) and the degree of yellow to blue (i.e. the **b** value with increasingly positive values approaching yellow and increasingly negative values approaching blue) were measured.

Table 9. Colour evaluation for Japanese imported soymilk

Variety	L	a	b
DH530 (Blue Tetra Pak)	78.85	2.56	3.33
Kinusayaka (White Tetra Pak)	79.81	1.77	6.78

The colour difference, or ΔE , between two colours (i.e., L1a1b1 and L2a2b2) is a valuable tool that can be used to determine color difference. A ΔE value of 1.0 is assumed to be barely perceptual to a trained eye (Source: http://www.newsandtech.com/issues/2005/06-05/pt/06-05_nate.htm, Spectrophotometers and Delta-E: Your color ruler, June 2005, by John Nate).

The ΔE value for Japanese soymilk was determined to be 3.67, indicating that a trained eye may be able to perceive a difference between the samples.

Sensory Evaluation of Soymilk Prepared at GFTC using Five Varieties of Soybeans

Mean scores for the trained panel evaluation of each sensory property of the soymilks produced by GFTC are presented



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in Table 10. Significant differences were observed among the five samples for sweetness. OAC Champion was significantly less sweet than Kinusayaka and S03W4. No differences in intensity were observed for the attributes of raw green, cooked bean, bitter and astringent.

Table 10. Mean scores and Tukey's HSD results for sensory properties of GFTC produced soymilks

Attributes		Samples				
		V	W	X	Y	Z
Raw Green	Mean	3.1a ^{1,2}	3.6a	3.8a	3.5a	3.8a
	St Error	0.27	0.32	0.31	0.32	0.35
Cooked Bean	Mean	5.4a	6.1a	5.2a	5.2a	5.3a
	St Error	0.33	0.33	0.37	0.32	0.32
Sweet	Mean	3.2c	4.2ab	3.7bc	3.9abc	4.5a
	St Error	0.35	0.34	0.30	0.37	0.29
Bitter	Mean	3.1a	2.5a	2.7a	2.8a	2.4a
	St Error	0.35	0.32	0.34	0.31	0.29
Astringent	Mean	3.0a	2.9a	3.1a	3.0a	3.0a
	St Error	0.31	0.36	0.30	0.28	0.32

¹ Means in a row with the same letter are not significantly different (P<0.05)

² n=38

Sample V – OAC Champion

Sample W – Kinusayaka

Sample X – DH618

Sample Y – DH530

Sample Z – S03W4

Colour Evaluation of Soymilk Prepared at GFTC using Five Varieties of Soybeans

Table 11 below provides the average score for colour evaluation of GFTC produced soymilks.

Table 11. Colour evaluation for GFTC produced soymilks

Variety	L	a	b
OAC Champion	78.70	2.01	7.83
Kinusayaka	77.67	2.09	5.30
DH 618	77.22	2.48	7.28
DH530	78.13	2.47	5.43
S03W4	78.62	2.09	7.06

The ΔE value was also determined between GFTC produced soymilk samples. Based on the ΔE value, a perceptual difference between soymilk samples OAC Champion-S03W4 as well as between soymilk samples Kinusayaka-DH530 is unlikely as the ΔE was below 1.0. All other comparisons had a value above 1.0, but below 2.7, indicating that a trained eye may be able to perceive a difference. Refer to Table 12 for a summary of ΔE comparisons between soymilk varieties.



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Table 12. Colour difference, ΔE , between soymilk varieties produced at GFTC

	OAC Champion	Kinusayaka	DH618	DH530	S03W4
OAC Champion		2.7	1.7	2.5	0.8
Kinusayaka			2.1	0.6	2.0
DH618				2.1	1.5
DH530					1.7
S03W4					

In summary, sensory evaluation results of the Japanese imported soymilks indicated a significant difference in the attributes of raw green, cooked bean, sweet and bitter between the samples. However, no differences in astringency were observed. Based on the colour measurements, a perceptual difference may exist between the two imported Japanese soymilk samples. Sensory evaluation of the GFTC-produced soymilks indicated significant differences among the five varieties for sweetness. OAC Champion was significantly less sweet than Kinusayaka and S03W4. No differences in intensity were observed for the attributes of raw green, cooked bean, bitter and astringent. Based on the colour measurements, a perceptual difference may exist between the soymilk samples with the exception of the OAC Champion/S03W4 and the Kinusayaka/DH530 samples.

Results and Discussion - Tofu

Sensory Evaluation of Tofu Produced at GFTC

Mean scores for the tofu are shown in Table 13. Significant differences were observed between Sample U and Sample X for beaniness. Sample X was perceived to have a significantly lower beany note than Sample U. No other significant differences in sensory properties were observed among the samples.

Table 13: Mean scores and Tukey's HSD results for sensory properties of GFTC manufactured tofu

Attributes		Samples					
		U	V	W	X	Y	Z
Bean	Mean	5.7a ^{1,2,3}	5.0ab	5.1ab	4.5b	5.1ab	5.0ab
	St Error	0.29	0.33	0.28	0.28	0.33	0.30
Sweet	Mean	1.6a	1.7a	1.7a	1.8a	1.9a	1.7a
	St Error	0.24	0.25	0.24	0.25	0.31	0.21
Bitter	Mean	2.7a	2.2a	2.2a	2.4a	2.4a	2.4a
	St Error	0.36	0.31	0.28	0.35	0.28	0.27
Astringent	Mean	3.8a	3.6a	3.7a	3.3a	3.9a	3.4a
	St Error	0.36	0.33	0.36	0.33	0.31	0.27

¹ Means in a row with the same letter are not significantly different (P<0.05)

² n=34

³ All data input on a 10 cm line where 0=low, 5= mid, and 10=high

Sample U = OAC Champion

Sample V = S03W4



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Sample W = DH530
Sample X = DH410SCN
Sample Y = DH420
Sample Z = Stargazer

Tofu Colour Evaluation

Table 14 below provides the average scores for colour evaluation, where the lightness (i.e., the **L** value expressed as 0 for black to 100 for white), the degree of red to green (i.e. the **a** value with increasingly positive values approaching red and increasingly negative values approaching green) and the degree of yellow to blue (i.e. the **b** value with increasingly positive values approaching yellow and increasingly negative values approaching blue) were measured.

Table 14. Colour Evaluation for GFTC-Produced Tofu

Variety	L	a	b
OAC Champion	89.4	3.2	10.6
S03W4	90.1	3.1	9.3
DH530	87.2	3.8	6.9
DH410SCN	90.0	3.1	8.4
DH420	89.2	3.5	8.8
Stargazer	89.0	3.3	8.7

The colour difference, or ΔE , between two colours (i.e. $L1a1b1$ and $L2a2b2$) is a valuable tool that can be used to determine colour difference. A ΔE value of 1.0 is assumed to be barely perceptual to a trained eye (Source: http://www.newsandtech.com/issues/2005/06-05/pt/06-05_nate.htm, Spectrophotometers and Delta-E: Your color ruler, June 2005, by John Nate).

Based on the ΔE value, a perceptual difference between tofu samples Stargazer-DH420, DH410SCN-S03W4 and DH410SCN-DH420 is unlikely as the ΔE was 1.0 or below. All other comparisons had a value above 1.0, but below 4.4, indicating that a trained eye may be able to perceive a difference. Refer to Table 15 for a summary of ΔE comparisons between tofu varieties.

Table 15. Colour difference, ΔE , between tofu varieties produced at the GFTC

	OAC Champion	S03W4	DH530	DH410SCN	DH420	Stargazer
OAC Champion						
S03W4	1.5					
DH530	4.4	3.8				
DH410SCN	2.3	0.9	3.3			
DH420	1.9	1.0	2.8	1.0		
Stargazer	2.0	1.2	2.6	1.1	0.3	

Tofu Texture Evaluation

Table 16 and Figures 6 and 7 below compare the firmness for the six tofu varieties.



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Table 16. Comparison of firmness values for six samples of block tofu

Sample	Firmness (g)			
	Day 1*	Day 3*	Day 5*	Compiled†
A	200 ± 50.9 ^b	221 ± 39.0 ^b	232 ± 30.1 ^c	217 ± 40.8 ^b
B	212 ± 65.8 ^b	252 ± 66.8 ^{ab}	236 ± 51.0 ^{bc}	234 ± 60.3 ^b
C	388 ± 114 ^a	367 ± 104 ^a	394 ± 110 ^a	383 ± 103 ^a
D	245 ± 60.3 ^b	275 ± 56.6 ^{ab}	273 ± 62.0 ^{abc}	264 ± 57.8 ^b
E	331 ± 60.2 ^{ab}	323 ± 71.5 ^{ab}	361 ± 62.3 ^{ab}	338 ± 63.1 ^a
F	220 ± 50.2 ^b	206 ± 44.7 ^b	209 ± 65.9 ^c	212 ± 51.4 ^b

Values with different superscripts in a column are significantly different ($p \leq 0.05$).

* n = 6

† n = 18

Sample A = OAC Champion

Sample B = S03W4

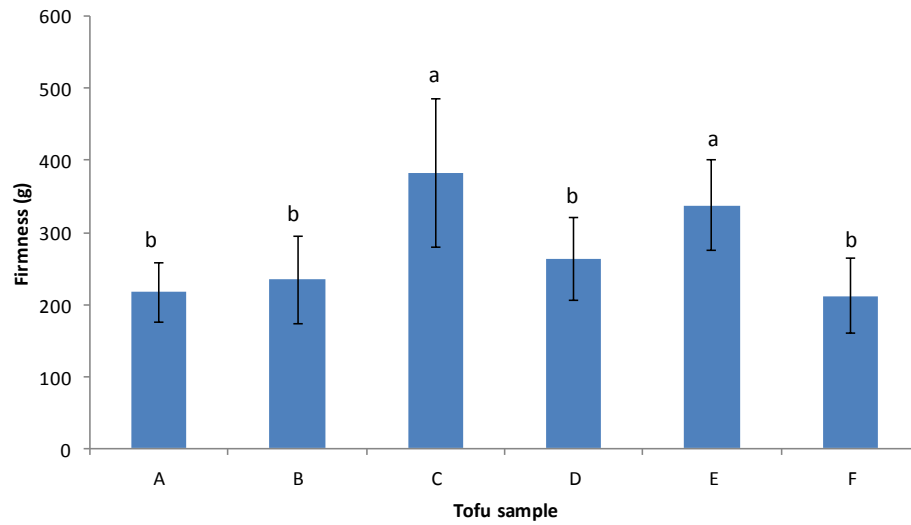
Sample C = DH530

Sample D = DH410SCN

Sample E = DH420

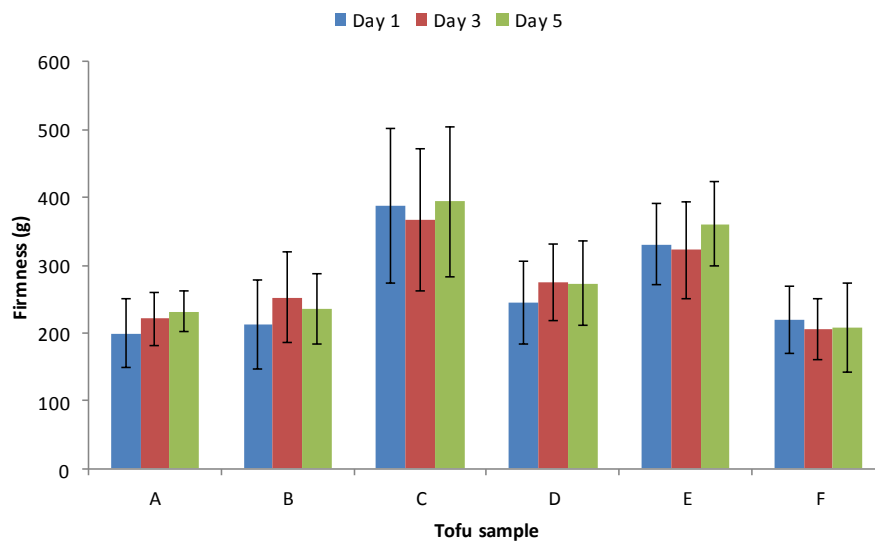
Sample F = Stargazer

Figure 6. Compiled firmness values for six samples of block tofu during five days of storage.



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Figure 7. Comparison of firmness values for six samples of block tofu during five days of storage.



Variability in tofu firmness was influenced by the inhomogeneity of the soybean curd structure. Obvious surface structural defects were avoided when arranging test blocks for texture analysis. However, concealed internal inhomogeneity could not be avoided. Softer samples (e.g. samples A and F) exhibited greater moisture expulsion during testing than firmer samples (e.g. samples C and E). Softer samples also exhibited a larger number of surface and internal structural discontinuities (i.e. open pockets or breaks in the curd). Although not significant, the different tofu samples exhibited three rough groups of firmness: samples C and E were consistently the two firmest samples, followed by the mid-range firmness of samples D and B. Samples A and F were consistently the least firm of all the samples tested.

In summary, sensory evaluation of the GFTC-produced tofu indicated a significant difference between OAC Champion and DH410SCN for beaniness. Based on the colour measurements, a perceptual difference may exist between the tofu samples with the exception of Stargazer-DH420, DH410SCN-S03W4, and DH410SCN-DH420 samples. Although not significant, the different tofu samples exhibited three rough groups of firmness. Samples DH530 and DH420E were consistently the two firmest samples, followed by the mid-range firmness of samples DH410SCN and S03W4. Samples OAC Champion and Stargazer were consistently the least firm of all the samples tested.

Results and Discussion – UPLC MS Q-TOF Analysis

Determine what components of the soybean, once processed into foods, are responsible for specific sensory responses and validate the identified soybean components as being pivotal in delivering the desired sensory evaluation. Using this information, recommend new varieties that contain the essential compounds for testing by the end user and sensory evaluation.

Six varieties were used to make tofu that was evaluated for taste by a sensory panel. In the tofu taste panel, the beany

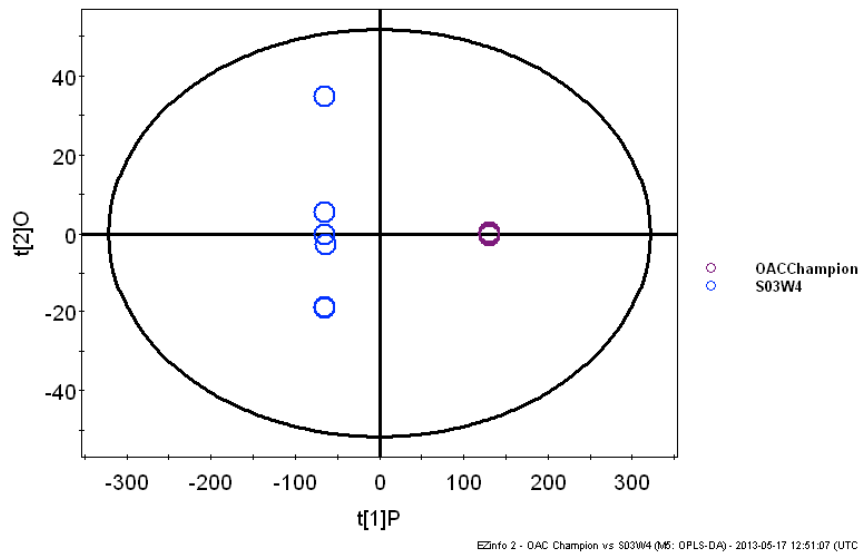


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taste attribute was statistically significant. Within the beany attribute, the difference between OAC Champion and DH410SCN was statistically significant (Table 13). These two varieties were subjected to metabolomic analysis. Principal component and discriminant analysis was then performed to identify the markers that are responsible for the attributes that show statistically significant taste results obtained from the GFTC sensory evaluation report.

In soymilk, varieties OAC Champion and S03W4 were different in one attribute (sweetness) and the other attributes were not significantly different (Table 10). Similar placement of the varieties in scores plot was obtained when grouping was performed based on four attributes. Two batches of S03W4 and OAC Champion were compared within the attribute of sweetness (Figure 8). Discriminant analysis for sweetness of OAC Champion, S03W4 revealed biomarkers. The following markers were discriminant (Figure 9) in S03W4: 254.9581 at 3.88 minutes, 254.9581 at 3.31 minutes, 270.9500 at 3.31 minutes, 270.9499 at 4.33 minutes and 295.1296 at 2.59 minutes. Table 17 reports on the identification of these biomarkers based on retention time and accurate mass.

Figure 8: PCA of two batches of S03W4 and OAC Champion were compared based on differing sweetness profiles in soymilk.





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Figure 9: Discriminant analysis of two batches of S03W4 and OAC Champion soymilk

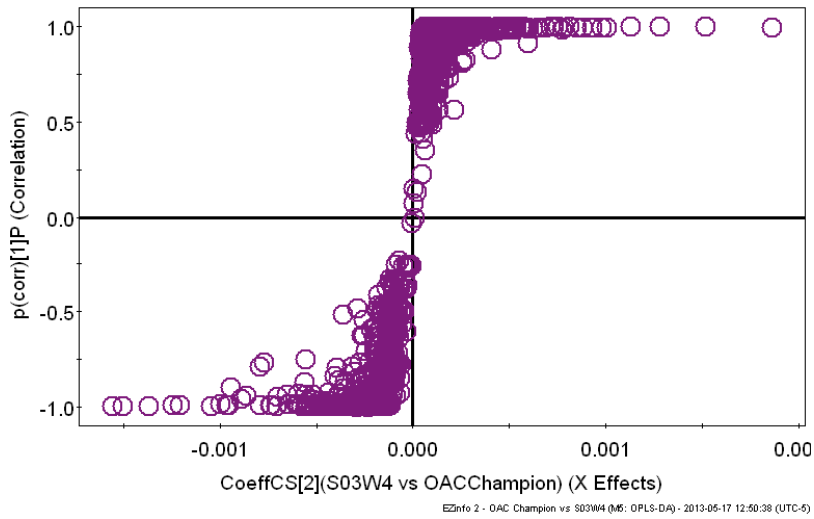


Table 17. Identification of discriminant biomarkers for sweetness in soymilk between OAC Champion and S03W4

Primary ID	Retention time (min.)	Mass (atomic mass unit)	S03W4	OAC Champion	Factor of change	Uncertainty
3.88_254.9581	3.88	254.9581	432.77	0.00554767	10000.0	100.000
3.31_254.9581	3.31	254.9581	307.121	0.0071708	10000.0	10.000
3.80_270.9500	3.8	270.95	359.93	0.0143433	10000.0	10.000
4.33_270.9499	4.33	270.9499	467.995	0.00405671	10000.0	105.000
2.52_295.1296	2.52	295.1296	0.0326648	631.921	10000.0	0.000
2.52_295.0139	2.52	295.0139	258.322	0.450836	573.0	271.782
1.33_311.1242	1.33	311.1242	0.00526544	416.004	10000.0	0.000
4.32_519.1136	4.32	519.1136	0.034572	296.542	8577.5	0.000

For Tofu, Figure 11 identifies the discriminant markers present while comparing DH410SCN and OAC Champion and Table 18 reports their identification based on retention time and accurate mass. The biomarker at Rt=6.9, m/z= 941.295 was shown to lower beany flavour.



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Figure 10: PCA analysis of DH410SCN and OAC Champion. Grouping is based on beany attribute in tofu.

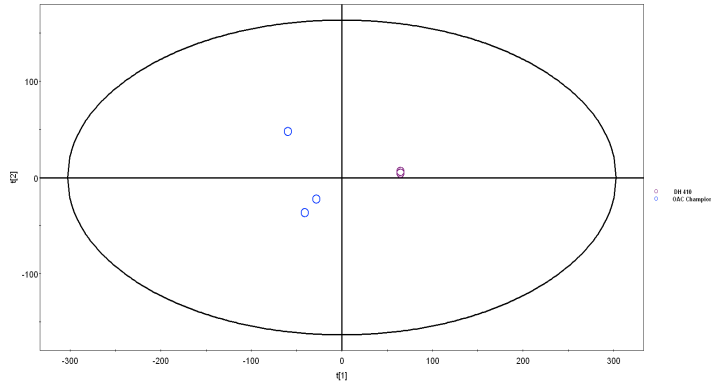
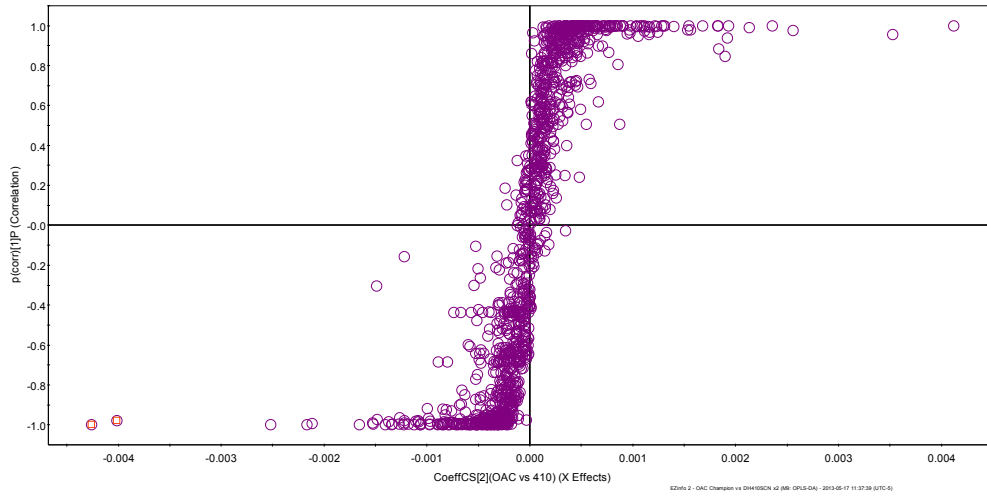


Figure 11: Discriminant analysis of OAC Champion versus DH410SCN





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Table 18. Identification of discriminant biomarkers of OAC Champion and DH410SCN (beany taste in tofu)

Primary ID	Retention time (min.)	Mass (atomic mass unit)	OAC	410	Factor of change	Uncertainty
6.61_943.3095	6.61	943.3095	68.45	294.795	4.3	0.117
6.90_941.2950	6.9	941.295	0.0150085	304.828	10000.0	0.000
6.61_943.7084	6.61	943.7084	298.53	0.0562382	5308.3	10000.000
6.98_1069.7816	6.98	1069.782	335.329	8.09296e-006	10000.0	10000.000

Compositional comparison was performed on 21 popular commercial varieties to determine if Sevia International currently has varieties that match these popular, commercially available soybean varieties and, thus, should provide similar end product results. Matches of each of these 21 popular commercial varieties were found using 34 of Sevia International's commercial varieties and 17 of Sevia International's experimental varieties.

Results and Discussion – Japan Collaborators’ Input into Sensory Studies

All three Japanese collaborators confirmed that the processes (both soymilk and tofu) used at GFTC for sensory evaluations were similar to that used in Japan. First, panelists are recruited and then tested to see if they can differentiate between samples (i.e. the triangle test), followed by training on various attributes and then they progress through sensory evaluations.

In addition to the traditional evaluation techniques used in this study, it was learned that the Japanese also give an overall rating of the food product based on the whole experience of consuming the product. This experience was best described by a tofu quality control specialist. It was explained that “humi” is a Japanese word, which refers to overall taste and smell of a food product. When the Japanese evaluate a food product, 60% of the overall experience comes from the texture and 40% of the experience comes from the humi. Texture is very important to the Japanese culture. Although, this overall experience would not have benefited this particular activity it was an interesting note to consider moving forward for future projects. An example of this type of rating system can be seen below in the results from the tofu testing done in Japan (Table 19). All factors are taken into consideration to provide a letter grade/rating. When evaluating tofu for humi, the tofu quality control specialist evaluates the smell of each tofu first and arranges them from weakest smell to strongest smell, he then proceeds to taste the weakest smelling tofu, followed by the second weakest smelling tofu. After evaluating these two samples individually, he picks his overall preference and only that sample moves on to be compared to the following sample. Generally, three samples are compared per sitting.

Also noted as a result of this activity was that although the lexicon words were appropriate and similar to those used in Japan, there was variance across perception of the attributes during the evaluations. For example, the Japanese collaborators found sweetness easily detectable in both the soymilk and tofu samples while this attribute was a challenge for Canadians to detect in the tofu samples. Figure 12 and Figure 13 compare a sensory evaluation ballot from Nadia, project manager at the GFTC, and a tofu quality control specialist, respectively. With the exception of the attribute astringency, the tofu quality control specialist has a broader range of quantifications for each attribute. This was consistent across other Japanese collaborators as well. Although the range varied over attributes between the Japanese collaborators and the GFTC panel, the Japanese collaborators were satisfied that the Canadian panel results would be acceptable in identifying constituents for biomarker identification.

Brenda Simmons 13-5-30 7:46 PM
Comment [4]: This is why AAFC wanted Japanese panellists. Caution!



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Figure 12: Sensory evaluation ballot from Nadia, project manager at the GFTC



check breakout note

Product: Tofu Product ID: _____

Judge # _____

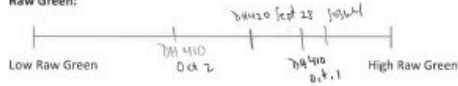
Name: NADIA

Date: Oct. 2/12

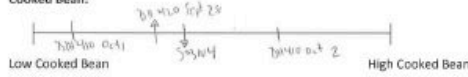
Please evaluate the attributes below for tofu. Make a vertical line on the horizontal line to indicate your rating of the attribute.

So3 n4 - sulfury.

Raw Green:



Cooked Bean:



Sweet:



Bitter:



Astringent:



Ferrous 410 Oct 2 sulfury



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Figure 13: Sensory evaluation ballot from Japanese tofu collaborator



Product: Tofu

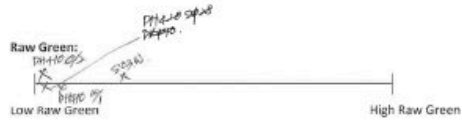
Product ID: _____

Judge # _____

Name: _____

Date: Dof. 2010

Please evaluate the attributes below for tofu. Make a vertical line on the horizontal line to indicate your rating of the attribute.





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Results and Discussion – Japanese Tofu Testing

Sevita International sent 36 samples of their various soybean varieties (including duplicate and triplicate reference samples) to a Japanese tofu testing laboratory for analysis to determine potential interest for each variety by tofu manufacturers. The results from this evaluation can be seen in Table 19. To date, Sevita International has sent varieties to approximately 14 companies based on the recommendations from this research as well as the information listed in Table 19.

Table 19. Japanese tofu evaluation results

Sample Name	Variety Name	Result
EXP10ENT23	DH401	B
EXP38ENT22	DH401	B
EXP38ENT22	DH401	C
EXP10ENT26	DH408	C
EXP10ENT03	DH863	B
EXP10ENT40	DH863	C
EXP40ENT17	HS05-03/a/1/1/1/21c	B
EXP40ENT17	HS05-03/a/1/1/1/21c	B
EXP39ENT20	HS05-18/1b/1/1/1/1a	B
EXP39ENT04	HS05-27/1/1/1/1a	C
EXP10ENT09	Leo	A
EXP10ENT41	Misty	C
EXP10ENT42	PSX11C13P	D
EXP10ENT44	PSX11C15G	D
EXP10ENT46	PSX12C01G	C
EXP38ENT20	PSX12C52S	C
EXP42ENT01	PSX12C61S	C
EXP42ENT15	PSX12C62S	B

Sample Name	Variety Name	Result
EXP42ENT15	PSX12C62S	B
EXP42ENT08	PSX12C71S	A
EXP42ENT12	PSX12C72S	B
EXP42ENT17	PSX12C81S	B
EXP10ENT45	PSX12C82G	C
EXP43ENT02	PSX12C91S	B
EXP10ENT04	Savanna	B
EXP10ENT29	Stargazer	B
EXP39ENT20	SVX13&0S3	A
EXP39ENT08	SVX13T0S1	B
EXP39ENT07	SVX13T0S2	C
EXP40ENT08	SVX13T0S5	B
EXP40ENT08	SVX13T0S5	C
EXP40ENT20	SVX13T0S6	B
EXP40ENT20	SVX13T0S6	B
EXP40ENT14	SVX13T0S7	C
EXP40ENT14	SVX13T0S7	B
EXP10ENT43	Thames	C

A= Excellent, B= Good, C= Acceptable, D= Unacceptable

Conclusions and Next Steps

Sensory evaluations were successful in identifying significant differences between varieties being tested. Further studies are needed to increase the number of varieties being evaluated so that further significant differences can be identified and the biomarkers identified via discriminant analysis can be further validated.

Texture qualities would also be beneficial to add to future tofu evaluations as suggested by both Japanese tofu collaborators. Texture components to consider would be chewiness or smoothness. Texture like pudding is preferable as silken tofu is preferred in Japan – some resistance to bite, but smooth and soft after the bite breaks through. Smaller pores within the tofu are desirable and contribute to this mouth feel.



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Overall the activity was a success and has already been a benefit to the industry partner, Sevita International, as the results from the University of Ottawa allowed to make recommendations to customers as to new varieties that should enter into their production trials. Sevita International customers have been receptive to the suggestions and have shown interest in the techniques used to make these recommendations. Positive results from Japanese production tests would result in an increase in export seed sales, which would have a positive domino effect down the supply chain increasing demand for Canadian soybeans and increased opportunities for Canadian farmers.

B (I). Funded Collaborators (Co-PI, AAFC, other federal scientists)

- Include the name of scientist / organization.

GFTC
Xin Hu – Process Development Specialist
Nadia Brunello-Rimando – Project Manager, Product Development Services
Karen McPhee – Manager, Product Development Services

Japanese Collaborators
Confidential

Sevita International
David Hendrick, CEO and Chairman of the Board
John Hendrick, Export Sales Japan
Mark MacDuff, Trait Development Manager
Jim McCullagh, Vice President (Research)
Stacey Simpkin, Research Coordinator

University of Ottawa
D. John T. Arnason - Professor
Ammar Saleem – Research Associate
Rui Liu – Laboratory Technician

B (II). Acknowledgement of non-funded collaborators (who provide support, e.g. access to other laboratory or other facilities and equipment input / advice / guidance / assistance, etc).

- For research supported by targeted funding programs, list any collaborators who are receiving Contribution Vote 10 funds (e.g., university and industry collaborators). In addition, please list separately the participants who support your project but are not receiving any funding through the program.
- Include name of scientist / organization.

Mark Berhow, USDA, Peoria, IL – Soybean Saponin Collaborator
Philippe Seguin, MacDonald College of McGill, Phytochemistry Collaborator

C. Variance Report (if applicable, describe how the work differs from the proposed research)



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- Include changes to objectives and project work plan / budget, changes to the team, other constraints.
-
- No changes to the objectives or project work plan

- D. Impact Assessment** (if applicable, describe how the variance factors above will impact project continuation)
- Include changes to the objectives, changes to the project work plan / budget, changes to performance (i.e. meeting targets).
-
- No changes to the objective or project work plan

- E. Achievements** (include only those related to this project)
- Include innovations, publications / conferences, technology transfer, capacity building, success stories, media, recognition and other outputs.
-
- Major achievements include:
- A successful sensory panel was conducted with significant differences identified.
 - Elucidation of biomarker differences in soymilk and tofu with soybean primary material.
 - Method to identify biomarkers in preferred varieties of tofu and soymilk. Specific biomarkers were identified.
 - Recommendations for new varieties to test were made to Japanese customers of Sevita International using the information derived from this activity.
- An initial research paper on soybean metabolome is in preparation and other publications on applications are planned. All papers will feature Sevita International's germplasm and will be submitted for approval to the company before publication.

- F. Lessons learned** (self-evaluation of project)
-
- Metabolomics appears to be a very powerful, new scientific tool allowing identification of biomarkers for food and taste preference. There are challenges in sensitivity and machine and biological variation in the metabolome. Large batch comparison is essential to reduce the analytical variation. Larger sample size from the taste panels with greater statistical power would greatly improve results. Identification of markers was successfully achieved, but as they are newly identified compounds in soybean, elucidation of structures is expected to be very time consuming.



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Jim McCullagh	May 31 2013	
PI Name	Date	Signature

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