NutraScan Artificial Gut

... Fully Automated Enzymatic
Digestion System

Simulated Enzymatic Digestion System for use in studying nutrients in foods and animal feeds

Next Instruments

... Bringing you the Next generation of analysers

Automated Simulated Enzymatic Digestion

Today's highly processed foods are different to the foods that the human body has evolved to digest. The human digestion system is the same however the make up of the digest and how our bodies react to the digest are important for health and socio-economic reasons.

The NutraScan Artificial Gut has been developed in order to study how the human and animal body digest foods. NutraScan simulates the enzymatic digestion of food under a series of incubations, at physiological pH and temperature that essentially mimics the buccal, gastric and pancreatic phases of food digestion. Protein, fat and starch are sequentially hydrolysed using a combination of hydrolytic enzymes as are found in the mouth, stomach and small intestines.

The digest can then be sampled and injected into a number of detectors in order to analyse the digest composition. Detectors such as a Glucose analyser can be used for measuring Glycemic Load and Resistant Starch. A HPLC can be used for measuring vitamins, proteins, organic acids and lipids. And a UV/Visible Spectrometer can be used to measure other compounds broken down from the food.

End point measurements can be made after the appropriate incubation period has been completed. Or rate studies can be undertaken whereby samples are measured at set time intervals.

The NutraScan Artificial Gut offers:

- Fully programmable
- 20 sample capacity
- 4 enzyme additions
- 8 buffer additions
- Variable temperature heating block
- Built-in stirrer
- Automatic injection valve
- Remote injection pulse
- Optional refrigerated sample collector

The NutraScan system can be setup to run for 24 hours unattended and the data will be collected and results plotted automatically.

Technology Description

The NutraScan System is designed to sequentially load enzymes and buffers to a sample of food while being held at 37^0C and under gentle agitation.

The food samples are chopped using a Zyliss kitchen food chopper to simulate the chewing process in the mouth. The food sample is weighed and added to a 120ml sample cup.

The NutraScan System has a 20 sample heating block with magnetic stirrer bars to provide a constant temperature of 37^0C and gentle agitation of the digest.

Six peristaltic pumps are used to load buffers and other reagents.

Two precision syringe pumps are used to accurately load the enzyme mixtures into each sample cup.

An XYZ Autosampler is used to load each sample cup in the required sequence

A peristaltic pump and an auto-injection valve are used to collect samples of the digest from each sample cup and either load them into the refrigerated sample collector or inject them into a detection system.

The NutraScan software runs on a Windows platform and controls each step of the procedure.

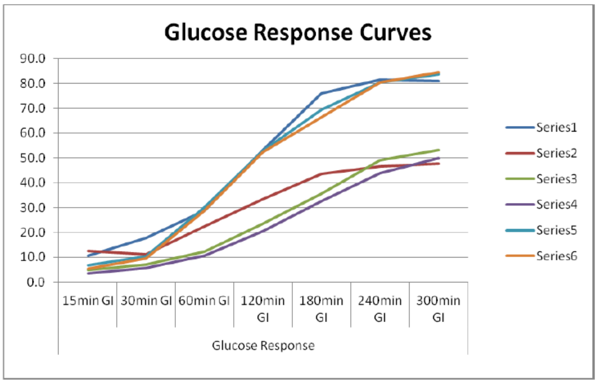
NutraScan Application Modules:

Predictive Glycemic Index:

The CSIRO's Division of Human and Animal Nutrition, Adelaide, SA, has developed an automated protocol for predicting Glycemic Index (GI) in cereal based foods.

Since cereal foods are the major source carbohydrates and thereby energy source for most humans, the CSIRO protocol is most appropriate for foods such as bread, biscuits, pasta, cake, breakfast products, rice and snacks.

The NutraScan Predictive GI Test has been adapted from the CSIRO protocol. A sample of food that contains an equivalent of 50mg of available carbohydrates is digested using buccal, digestive and pancreatic enzymes over a 5 hour period. Amyloglucosidase is then added to breakdown the starch molecules to glucose which is then measured using a Glucose analyser at 15, 30, 60, 120, 180, 240 and 300 minutes. The end point glucose concentration is used to compute the Predictive GI for the food sample.



$$\text{Predictive GI} = \frac{\text{Glycemic Response 50mg of Test Sample}}{\text{Available mg of Carbohydrate}} \times 100$$

Resistant Starch:

The digest remaining after the GI protocol is used to measure the Resistant Starch (RS) content of the food. Resistant Starch is the starch that is held inside the starch cells and is not broken down to form glucose in the small intestines. In the large intestines, Resistant Starch is fermented into small chain organic acids which are important for the endocrine system.

The digest is first filtered under vacuum to remove the glucose produced in the GI protocol. The cell walls are disrupted using concentrated Potassium Hydroxide under rigorous agitation. After rinsing and adjustment of the pH to 4.0, Amyloglucosidase is added to convert the starch to glucose which is then measured using the Glucose Analyser. The Resistant Starch is computed as the mass of the glucose generated after the cell walls have been disrupted. This mass is expressed as a percentage of the original sample weight.

Total Dietary Fibre:

The measurement of Total Dietary Fibre is a complex and difficult procedure. The enzymatic digestion steps are similar to the protocol used for measuring Resistant Starch.

The NutraScan Artificial Gut can be used to automate several of the steps required for measuring TDF. The initial digestion with alpha amylase at 90^{0}C is performed external of the NutraScan. This digest can then be added to the NutraScan reaction cups where the addition of buffers, other enzymes, rinsing and filtration can be performed automatically.

The residual is then removed, weighed and the protein and ash measurements performed to determine the TDF by difference.

Micro Nutrients:

During the enzymatic digestion of foods, micro nutrients are broken down. Proteins are converted to amino acids, fats to fatty acids and carbohydrates to simple sugars. As well vitamins and other compounds can be affected by the digestion process.

The NutraScan Artificial Gut provides a system where the progress of these nutrients can be monitored.

The NutraScan Artificial Gut can be connected to a number of analytical instruments whereby an extract from the digest can be collected and an analysis for specific compounds can be made.

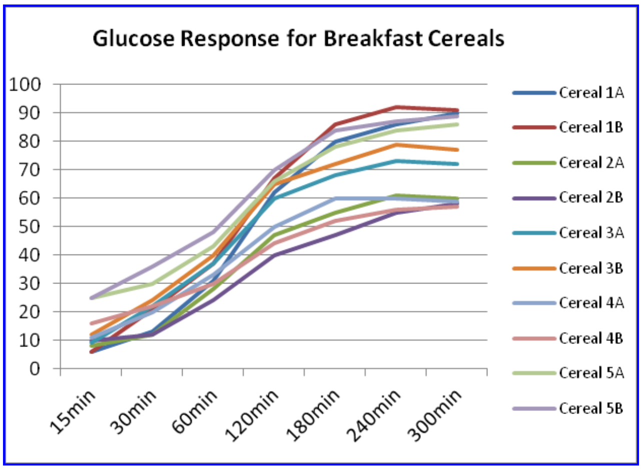
The NutraScan includes an auto injection valve and an external trigger that can be used with a Glucose Analyser, a HPLC system, a UV-Visible Spectrometer or even a LCMS system.

A Refrigerated Sample Collection Block is available to hold the digest samples which may be temperature labile.

In Vitro vs In Vivo Measurements:

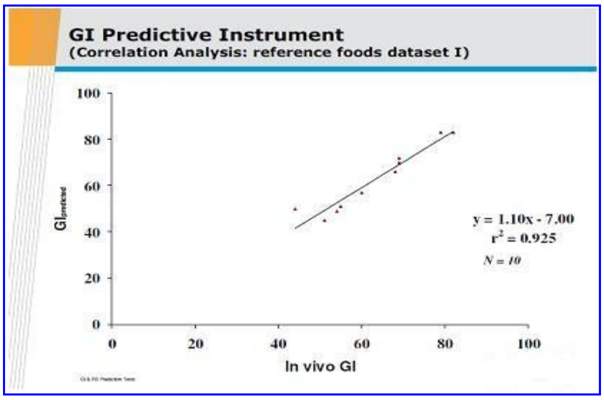
Glycemic Index is a measurement that is by definition an In Vivo measurement. It is an assessment of how humans digest carbohydrates in food and the rate of release of glucose into the blood stream. Glycemic Index is a measure of glucose in the blood stream over a 2 hour period. The blood glucose level is also regulated by insulin and gastric emptying. The stomach is continuously moving the food through the digestive tract and so the blood glucose level is reduced over time due to gastric emptying.

In Vitro measurement of Glycemic Index does not include the affects of insulin nor gastric emptying. The Predictive GI value obtained from the NutraScan GI Module is really a measure of the amount of glucose released from a food under simulated conditions found in the gut over a five hour period. The Predictive GI is based on the end concentration of the glucose in the digest, not a rate of increase in the blood glucose level. For most cereal based foods, the In Vitro Predictive GI value is highly correlated with the In Vivo GI method as are shown in the following plots. However many other foods, especially those containing added sucrose, fructose and high fat levels, require a correction equation to relate the In Vitro method to the In Vivo method.



The most significant advantages of the NutraScan Predictive GI and RS measurements are in the reduced cost of measurement and the higher precision of analyses. The diagram above shows the plots of the Glucose Response for several breakfast cereals analysed in duplicate. The Standard Deviation of Differences between duplicates is 3.1 units. For cereal based food, i.e., bread, plain biscuits, pasta, noodles, rice etc, the Predictive GI and RS methods correlates well with the In Vivo methods.

The following plot shows a comparison between the In Vitro GI and RS method vs the In Vivo methods for several cereal based foods.



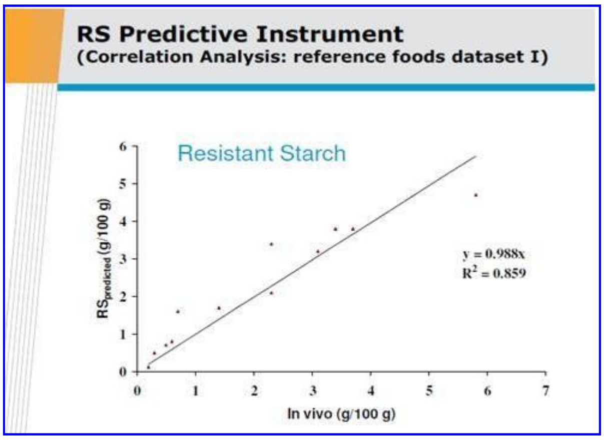


Table 1. provides a comparison between the In Vitro and In Vivo methods for both GI and RS.

Table 1 Comparative performance parameters for in vivo testing methods and concept RS and GI prediction instruments

Parameter	In vivo Testing Methods[1]		Prediction Instruments	
	GI	RS	GI	RS
Time required to produce results[2]	7 d	24 d		
Cost per test[3] ($USD)	$4,000	$8,000	$20-100	$20-100
Sample throughput[4] (test foods/week)	2	1-2	100-200	50
Discriminatory power[5]	15 GI units	0.5 g RS/g food	5 GI units	0.2 g RS/g food
Inter-Assay Precision (CV)[6]	30-40%	20-45%	1-2%	5-10%

NextInstruments

Next Instruments Pty Ltd
B1, 366 Edgar Street, Condell Park, NSW, Australia, 2200
Tel: 612 9771 5444, fax: 612 9771 5255
Email: sales@nextinstruments.net
Web: www.nextinstruments.net