

PRODUCT FOCUS



Gluten Analysis

Next Generation Gluten
Detection Technology



- ▶ **Next generation antibodies**
- ▶ **Proprietary methods based on G12 and designed for the food industry**
- ▶ **Targets the most toxic proteins**
- ▶ **Links celiac disease to food analysis**

Romer Labs® gluten tests use proprietary gluten detection methods, which employ a **next generation antibody**, called G12. The method represents a **quantum leap in gluten detection** as it **targets the most immunotoxic proteins** for those intolerant to gluten. This approach should be considered to be closer to the ideal of a food safety test as it establishes the important **link between celiac disease and** the detection of the immunotoxic peptides in **food**.

Romer Labs® test kits and services are specifically designed to meet the high standards of food manufacturers. Our validation criteria are among the strictest in our industry. Our test kits have been validated for all major food products. Our customers are in the food industry. We know their needs from our 30 years' experience in food safety diagnostics. We have our own facilities in seven countries to attend to the needs of multinational companies 24 hours a day around the globe. We operate accredited full-service labs on three continents, supporting our customers with fast turn-around times and cutting-edge instrumental analytical technology.

We are the experts in gluten analysis.



**AgraQuant® Gluten G12
ELISA**



**AgraStrip® Gluten G12
Lateral Flow Test**

- ▶ **Validated on all major food products**
- ▶ **Accredited full service laboratories**
- ▶ **Global network**
- ▶ **Unrivalled technical support and service**
- ▶ **30 years of experience in food safety diagnostics**

Tel : 02 8212 4074 Fax : 02 9423 6992
info@keydiagnostics.com.au
www.keydiagnostics.com.au
PO Box 1038, Gymea NSW 2227

keydiagnostics



Making the World's Food Safer®



AgraQuant® Gluten G12 ELISA

The AgraQuant® Gluten G12 assay is a sandwich enzyme-linked immunosorbent assay (ELISA). Gluten is extracted from a sample with the Romer® Extraction Buffer. The monoclonal G12 antibodies directed against the toxic fraction of gluten are pre-coated on the surface of a microwell. The extracted sample or standards are applied to the wells and the gluten binds to the antibodies. After a washing step, an enzyme-conjugated monoclonal antibody specific to the Gluten G12 protein is applied to the well and incubated. After a second washing step, an enzyme substrate is added and a blue colour develops. The intensity of the colour is directly proportional to the concentration of Gluten G12 in the sample or standard. A stop solution is then added which changes the colour from blue to yellow. The microwells are measured optically using a microwell reader with a primary absorbance filter of 450nm (OD₄₅₀). The optical densities of the samples are compared to the ODs of the standards and an interpolated result is determined.

Performance Characteristics

Limit of Detection: 2 ppm gluten
 Quantitation Range: 4 - 200 ppm gluten
 Covers all limits of Codex



Recovery

Matrix	Spiking level [ppm]	Concentration [ppm]	Recovery [%]
Curry Sauce	5	6.1	124.2
	20	21.1	105.5
Crisps	5	4.2	84.0
	10	8.1	81.0
	20	18.1	90.5
Chocolate (+ gelatine)	5	5.0	100.0
	20	16.3	81.5
Corn Snack	5	5.4	108.0
	10	8.5	85.0
Rice Flour	5	5.0	100
	20	16.9	84.5
Yogurt	5	5.5	110.0
	20	20.8	104
Paprika	2	5.3	106.0
	10	10.1	101.0
	20	16.3	81.5



AgraStrip® Gluten G12 Lateral Flow Test

The AgraStrip® Gluten G12 Test Kit is an immune-chromatographic test for screening food samples for gluten. During the test, the sample reacts with the coloured conjugates (anti-gliadin 33mer monoclonal antibody – red-coloured microsphere) previously fixed to the test. This complex spreads by capillary action through the membrane of the test. Everything is included in the kit and no further equipment is necessary. The test is easy to use, fast and reliable.

It is possible to adjust the AgraStrip® Gluten G12 to **any cutoff level** by varying the amount of dilution buffer. The kit includes an additional guide for the use of AgraStrip® Gluten G12 at 5, 10 and 20 ppm. Variations in levels may occur depending on the food matrix being tested. Romer Labs® can support customers, meeting their special needs by validating individual matrices.

Performance Characteristics

3 different **cutoff levels**:
5, 10 and 20 ppm
Takes just minutes to perform



LOD in spiked commodities:

	AgraStrip® Gluten G12	Other Gluten LFD
Yogurt	5 ppm	>10 ppm
Dairy Free Spread	5 ppm	>10 ppm
Chocolate	10 ppm	>10 ppm
Curry Sauce	5 ppm	>10 ppm
Rice Flour	5 ppm	>10 ppm

Romer Labs® G12 Methods

Recent discussions about celiac disease have taken us from the concept of gluten detection to the detection of the relative toxicity of gluten for the food safety. The strongest response is directed towards an α 2-gliadin fragment that is 33 amino acids long and a principal contributor to gluten immunotoxicity. This so-called 33-mer is highly resistant to breakdown by digestive enzymes and is, therefore, a suitable molecule for use as an analytical marker. Homologues have been found in food grains that are toxic for celiac patients, but are absent in nontoxic grains. For this reason, the ELISA test AgraQuant® Gluten G12 and the lateral flow device AgraStrip® Gluten G12, which uses the G12 antibody, have recently been launched by Romer Labs® to specifically measure the most immunotoxic proteins for those intolerant to gluten and thereby take analysis in a new direction in assuring food safety.



The G12 antibody recognizes the QPQLPY peptide from the toxic fragment called 33-mer with peptide structure of LQLQFPQPQLPYQPQLPYQPQLPYQPQPF of the gliadin protein present in gluten. It was confirmed during validation studies that G12 does not give any false positive signals with soy and is, therefore, suitable for measuring gluten in products containing soy. There is also no cross reactivity to maize or rice.

There is an ongoing debate concerning the presence or absence of gluten in oats. Several publications conclude that certain oat varieties may cause an auto-immune response in celiac patients. During the validation of AgraQuant® Gluten G12 and AgraStrip® Gluten G12, positive and negative responses to oat varieties were observed. The positive results appear to be a specific reaction of the antibody with the toxic fragment, rather than a non-specific response. Therefore, the G12 antibody may shed new light on this debate. In regards to this, the Spanish Celiac Association has recently awarded the 6th National Prize for Research on Celiac Disease to a scientific team that used the G12 antibody to identify oat varieties containing low levels of gluten.

The results obtained from new immunochemical test systems based on the G12 antibody should be considered to be closer to the ideal of a food safety test as they establish the important link between celiac disease and the detection of the immunotoxic peptides.



References:

Structural basis for gluten intolerance in celiac sprue
Shan L, Molbergv Ø, Parrot I, Hausch F, Filiz F, Gray GM, Sollid LM, Khosla C. Science. 2002 Sep 27;297(5590):2275-9.

Toward the assessment of food toxicity for celiac patients: characterization of monoclonal antibodies to a main immunogenic gluten peptide.
Morón B, Bethune MT, Comino I, Manyani H, Ferragud M, López, MC, Cebolla A, Khosla C, Sousa C. | PLoS ONE. 2008 May 28;3(5):e2294.

Sensitive detection of cereal fractions that are toxic to celiac disease patients by using monoclonal antibodies to a main immunogenic wheat peptide. Morón B, Cebolla A, Manyani H, Alvarez-Maqueda M, Megias M, Thomas Mdel C, López MC, Sousa C., Am J Clin Nutr. 2008 Feb;87(2):405-14.

The Molecular Basis for Oat Intolerance in Patients with Celiac Disease
Helene Arentz-Hansen, Burkhard Fleckenstein, Øyvind Molberg, Helge Scott, Frits Koning, Gunther Jung, Peter Roepstorff, Knut E., A. Lundin, Ludvig M. Sollid, PLoS Medicine, 2004 - medicine.plosjournals.org

Romer Labs, Inc. - America · Union, MO, USA · T +1 636 583 8600 · F +1 636 583 2340 · E office@romerlabs.com

Romer Labs Diagnostic GmbH - Europe · Tulln, Austria · T +43 2272 615 33 · F +43 2272 615 33 13177 · E office-europe@romerlabs.com

Romer Labs Singapore Pte. Ltd. - Asia · Singapore · T +65 6631 8018 · F +65 6275 5584 · E salesasia@romerlabs.com