

## A SIMPLE DOT-ASSAY FOR THE DETECTION OF ANTI-GAD ANTIBODIES

Autoantibodies to the enzyme glutamate decarboxylase (GAD) are found in very high titers in Stiff-Man syndrome (SMS). Recently it has been shown that these antibodies are also present in other neurological syndromes, such as progressive encephalomyelitis, sporadic progressive ataxia and epilepsy (1). Anti-GAD antibodies are also associated with type 1 diabetes but titers are much lower. Additionally in SMS and allied diseases, anti-GAD antibodies react with linear epitopes, common to GAD65 and GAD67, allowing the use of ELISA or immuno-dot techniques for their detection (2).

We report here on a simple, commercially available dot-test, for the rapid and simple detection of anti-GAD65 and GAD67 antibodies. Reagents are produced by D-TEK (Mons, Belgium). Diluted serum is incubated for half an hour with a reagent strip containing immobilized human-recombinant GAD65 and GAD67. After washing, the strip is incubated with a human anti-IgG antibody conjugated to alkaline phosphatase. After a new washing step the strip is incubated with the substrate for 10 minutes a blue spot appears when anti-GAD antibodies are present.

The sera of twelve patients affected with SMS or encephalomyelitis with ataxia were tested in comparison with ELISA and RIA. A strong reactivity was observed on the immuno-dot for anti-GAD65 antibodies. Reactivity to GAD67 was also very strong in 10 of the 12 patients (Fig.1).

\* D-TEK is the manufacturer of the ALPHADIA DOT kits.

This new Immuno-dot assay represents a simple method for the rapid detection of high titer antibodies to GAD65 and GAD67 as seen in SMS and other neurologic diseases. Nevertheless it cannot be used for the detection of anti-GAD65 antibodies in diabetes 1 as these antibodies react only with conformational epitopes not accessible on the dot.

#### REFERENCES

1. Brown P, Mardsen CD : The Stiff-Man and Stiff-Man plus syndromes. *J Neurol* 1999; 246 : 648 – 652
2. Lohmann T, Hawa M, Leslie RDG et al : Immuno reactivity to GAD in Stiff-Man syndrome and type 1 diabetes mellitus. *Lancet* 2000; 356 : 31 - 35

René Louis Humbel (\*)

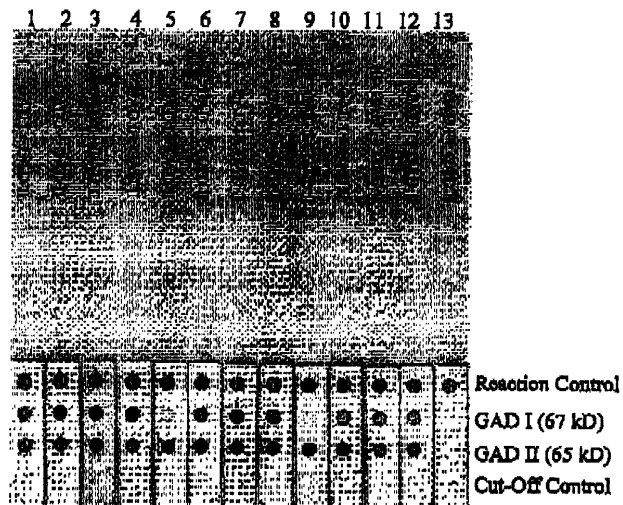
Patrick Schmit

Véronique Pierrard

Mireille Goeury

Georges Gilson

(\*) Immunopathology Lab, Centre Hospitalier de Luxembourg  
L-1210 Luxembourg



	Patient	GAD I (ELISA) N<7	GAD II (ELISA) N<1.5	GAD II (RIA) N<1.0
1	Laval.	94	139	14800
2	Bach.	294	321	23500
3	Foul.	746	700	93380
4	Kne.	302	277	24728
5	Mor.	28	160	18680
6	Bon.	188	317	40330
7	Hut.	231	456	38912
8	Port.	168	226	26710
9	Tab.	20	449	29000
10	Ras.	192	1207	61800
11	Ler.	48	594	4230
12	Stan.	86	253	23780
13	Negative Ctrl			