

Insecticide quality kits

Dr Derek Russell

NRI, University of Greenwich, UK
CESAR, La Trobe University, Melbourne
Australia

Components 7&8: Immunodiagnostic kits

	ELISA	DIP STICK		
Insecticide	Quality	Quality	Residue	Resistance
Pyrethroid	Done	Done	Done	?????
Endosulfan	Done	Done	Done	Prototype
Cry1Ac/1Ab	Comm.	Comm.	Done	Prototype
Cry1Ac+Cry2Ab	Done	Prototype	Done	Prototype
Carbamate	Done	Prototype	Prototype	Done
OP, Spinosad, Indoxacarb	Develop-ing	Develop-ing		Developing
HaNPV	Develop-ing			
Azadirachtin	Develop-ing			

Problems

- Insecticides are not proteins and therefore you can't raise antisera against them!
- Many molecules may-cross react (eg many pyrethroids)
- The level of specificity required determines the part of the molecule which should be used
e.g. do we want a kit for all phosphothiorinate OPs (chlorpyrifos, quinalphos, phoxim etc) or only one compound
- It is essential to be able to distinguish between the parent compound and its metabolites (possibly non-active)

Insecticide detection kits

Method:

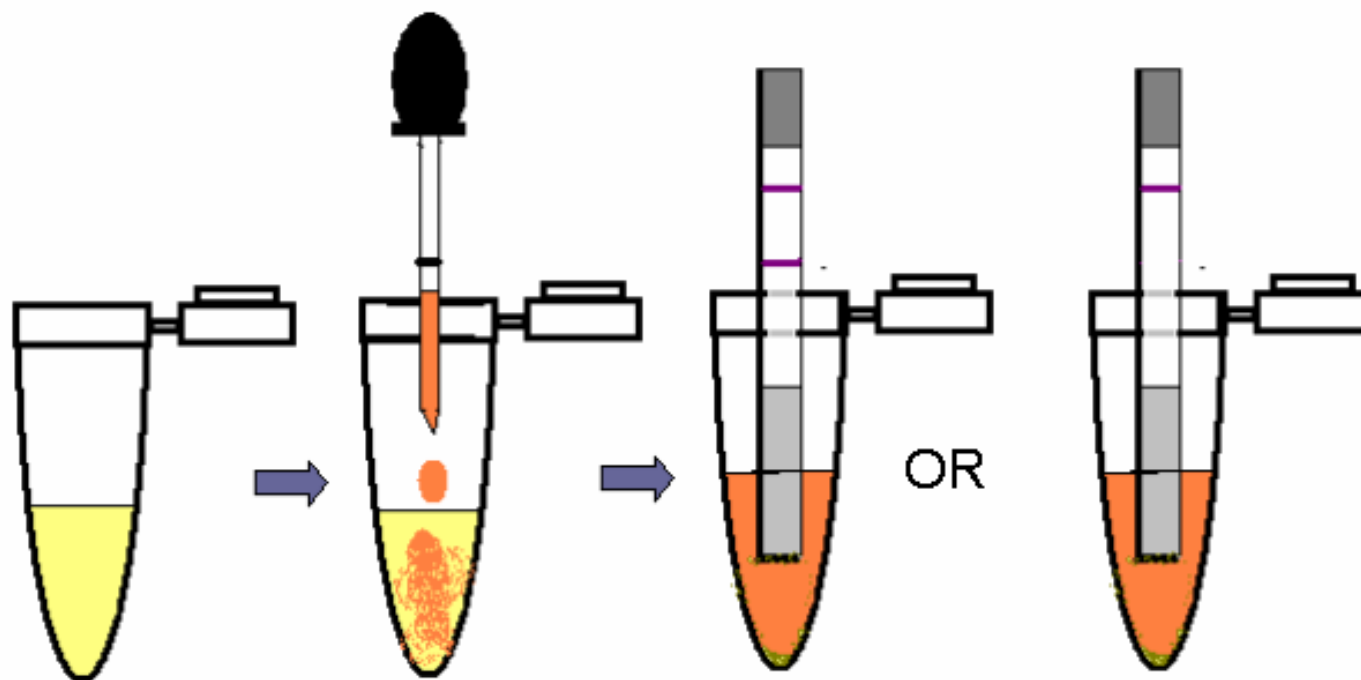
1. Conjugate the diagnostic part of the molecule with a protein (e.g. bovine serum albumen) to form a hapten
2. Inject the hapten into rabbits
3. Harvest the resulting antisera
4. Place the antisera conjugated with gold in the pad at the bottom of the strip
5. Place the antigen (**the chemical itself**) in a stripe further up the strip

The test for pesticide quality

1. Place the test insecticide in the vial.
2. Add the 'dip-stick'.
3. If there is not enough insecticide to immobilise it, the antibody moves up the strip and reacts with the antigen on the strip giving a 'bad quality sign'.
4. If there is at least enough insecticide, it reacts with the antibody on the 'pad', immobilising it. There is therefore no antibody to move up the strip and so no band.

Insecticide Quality Detection kit

Cypermethrin and Endosulfan



1. Buffer 0.5 ml

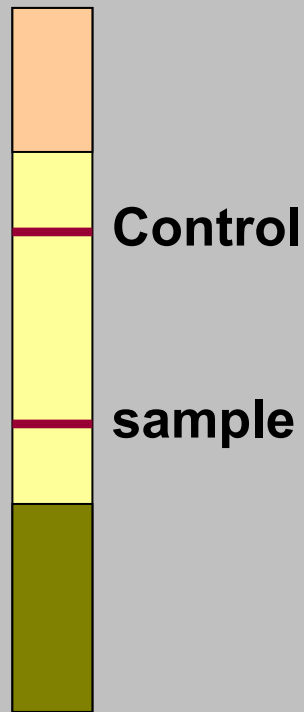
2. Add 0.01 ml insecticide

3. Poor quality

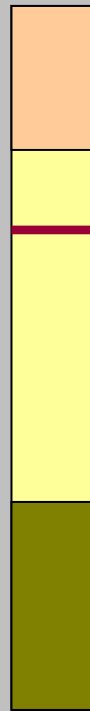
4. Good quality

The dip-stick strips (10 min)

Resistance detection



Positive



negative

Quality detection



positive



negative

Pesticide quality kits

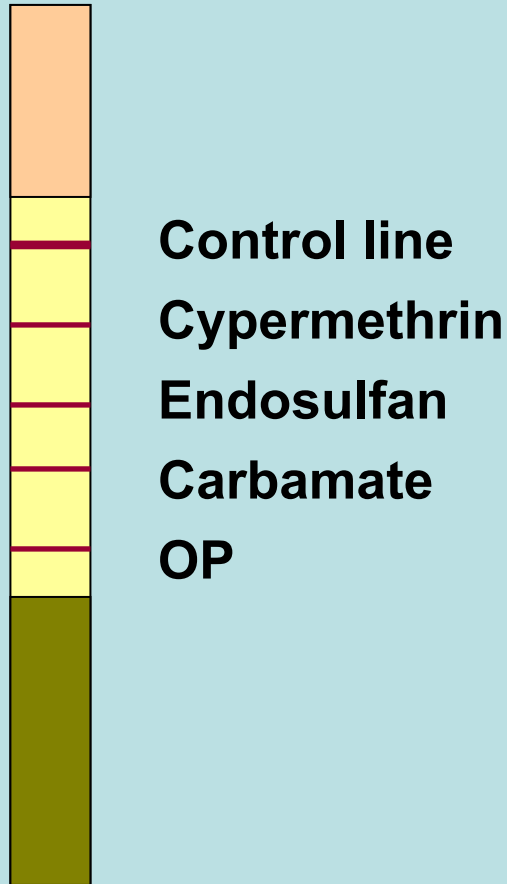
Imunodiagnostic:

- *Endosulfan* – unique to the molecule
- *Pyrethroid* – deliberately cross-reacting between cypermethrin, deltamethrin, fenvalerate and similar compounds

Phage display:

- Organophosphates
- Carbamates

The ladder strips



A single strip can be used to detect more than one toxin simultaneously

- **Diagnostic**
- **Quality detection**
- **Residues**

Example: Cry1Ac & Cry2Ab
(patent pending 600/del/2002)

Bt toxin quality kits

www.indiaagronet.com/innovativebio

Bt-TEST KITS

RAPID - RELIABLE - ECONOMICAL

IMMUNODIAGNOSTIC KITS

(Patent Pending)

Developed by :

CENTRAL INSTITUTE FOR COTTON RESEARCH
PANJARI, WARDHA ROAD, NAGPUR



Bt-Express

DIP STICKS

For Detection of
Cry 1 Ac/Cry 1 Ab in Transgenic Plants



Bt-Express

1. This is a dipstick format and can be used by even a layman.
2. Bt-Express has been designed for instantaneous detection of Bt- Toxin in either seeds or plant tissues.
3. It takes about 10 minutes for the test to be completed.
4. The test can be used in fields and does not require any additional facilities for use.
5. All material required for the testing is provided with the kit.
6. The kit is rapid, reliable and ready to use.

Quantitative ELISA

Detection sensitivity

BT:

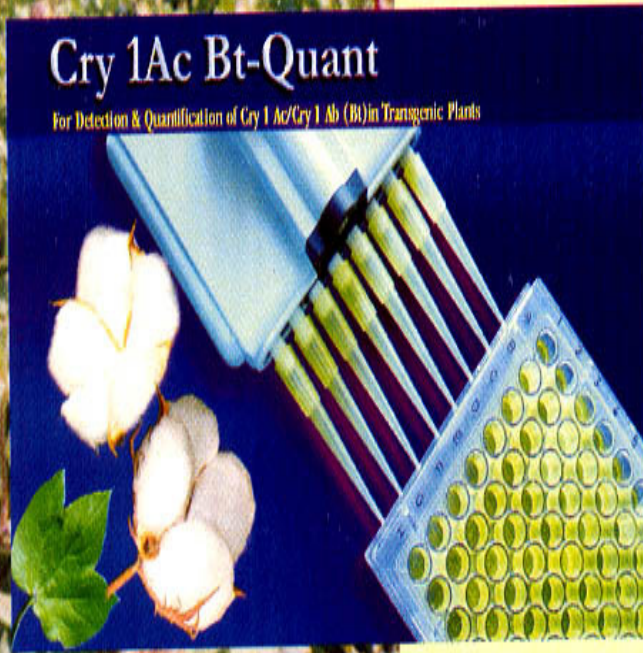
Cry1Ac: 10 ppb

Cry2Ab: 20 ppb

Conventional insecticides:

Pyrethroids: 2 ppb

Endosulfan: 5 ppb



Cry 1Ac Bt-Quant

1. The Cry1AC Bt -Quant is an ELISA kit, which facilitates a precise quantification of Cry1Ab or Cry1Ac, expressed in transgenic plants.
2. The kit is simple, cost effective and very reliable.
3. It takes about 2hrs for completion of one set of ELISA assay.
4. Each ELISA plate can be used for 96 samples (including four wells for standards and two for blank). Depending on the capabilities of a laboratory, hundreds of samples can be processed in a single day.
5. ELISA plate reader is a requirement for use of the kit.
6. Additionally the ELISA kit can also be used for the quantification of Bt-toxins in Bt(*Bacillus thuringiensis*) insecticide formulations. Separate standards will be provided on request.