DISEASE FREE SWEET POTATO SEED PRODUCTION

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Disease free sweet Potato seed

- No proper system for assessment of potato seed has been developed, However;
    - Should be free from all viral diseases
    - Should be either free from soil and tuber borne diseases or carry them within permissible limits
    - Should not have been grown in areas where weevils, nematode and other quarantine diseases are endemic

Sweet potato viruses

- Viruses are the most damaging group of disease organisms affecting sweetpotato in Africa.
- Transmission by vectors(aphid or whitefly)
- Once the virus enters a cell in the new host, forces the cell to produce more viruses identical to itself.

1.2. Recognized viruses that infect sweet potato

Genus	Virus	Transmission	Distribution
Potyvirus	SPFMV	Aphid	Worldwide
	SPLV	Aphid	Taiwan, China, Japan, Indonesia, Philippines, India, Egypt
	SPMSV	Aphid	Argentina, Peru, Indonesia, Philippines, China, Egypt, South Africa, Nigeria, New Zealand
	SPVG	Aphid	China, Japan, USA, Egypt, Ethiopia, Nigeria, Barbados, Peru, Spain, South Africa
	SPV2	Aphid	USA, Taiwan, China, South Africa, Portugal, Australia, Barbados
	SPCSV?	Unknown	Caribbean Region, Zimbabwe, Uganda, Kenya
	SPVMV	Aphid	Argentina
Ipomovirus	SPMMV	Whitefly?	Africa, Indonesia, China, PNG, India, Egypt, New Zealand
	SPYDV	Whitefly	Taiwan, Far East
Crinivirus	SPCSV	Whitefly	Worldwide

Sweet potato viruses in Kenya

- Two main sweetpotato viruses affect sweetpotato in Kenya;
  - Sweetpotato feathery mottle virus (SPFMV)) transmitted by aphids and
  - Sweetpotato chlorotic stunt virus (SPCSV) transmitted by whiteflies.
- Each virus by itself may cause only very mild symptoms but when a sweetpotato plant gets infected by both the viruses, a very severe disease results which is known as **sweetpotato virus disease** (SPVD).
- SPVD causes Yield looses up to 98%

Virus transmitters

Aphids
Aphis gossypii and others
(Homoptera: Aphididae)
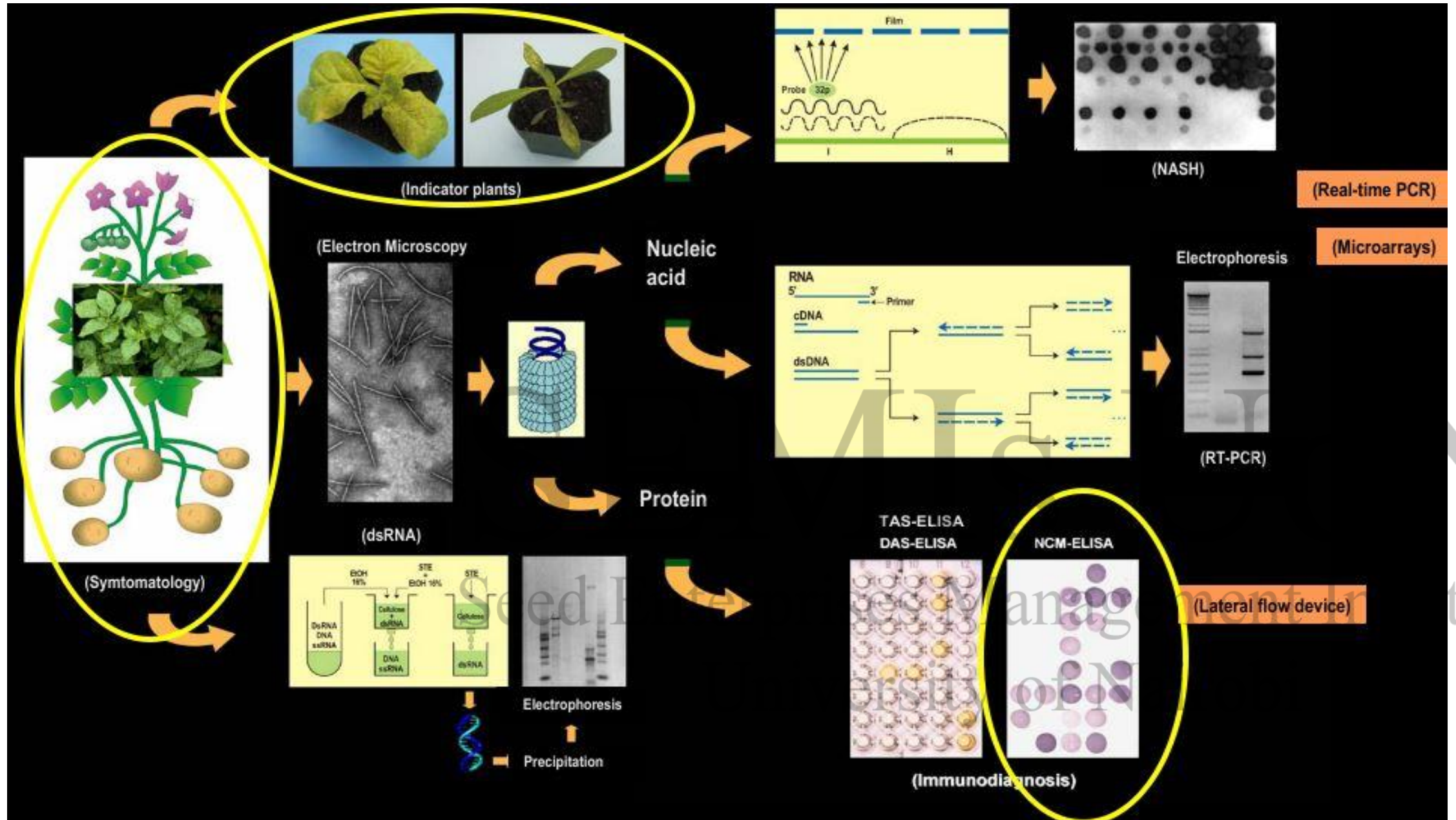

Whiteflies
Bemisia tabaci (Homoptera: Aleyrodidae)

Virus indexing

- Virus indexing combines knowledge of viruses with methodologies for their detection

- **Importance of virus indexing**

- To assure the effective safe movement of sweetpotato germplasm.

- **Challenges**

- low virus titers and

- absence of symptoms from single infections by most viruses

- **Remedy**

- grafting onto indicator plants to boost titers and detect viruses reliably.

- The commonly used indicator plant is *Ipomoea setosa* which is susceptible to most viruses infecting sweetpotato.

- **The procedure includes;**

- symptomatology in sweetpotato plants grown in greenhouse (pots),

- as well as in plants grafted with scions of the basal part of sweetpotato.

- Virus detection and identification is confirmed by serology with antisera available to known viruses.

2.0. Virus detection methods

(Indicator plants)

Film

Probe 32p

I H

(NASH)

(Real-time PCR)

(Microarrays)

(Electron Microscopy)

Nucleic acid

RNA
5' 3'
Primer
cDNA
dsDNA

Electrophoresis

(RT-PCR)

Protein

(dsRNA)

TAS-ELISA
DAS-ELISA
NCM-ELISA

(Symtomatology)

EtOH 18%
STE + EtOH 18%
STE

DsRNA DNA ssRNA
Cellulose + dsRNA
Cellulose

DNA ssRNA
dsRNA

Electrophoresis

Precipitation

(Lateral flow device)

(Immunodiagnosis)

2.1 Serological test

NCM-ELISA test

KIT

SPFMV
SPMMV
SPLV
SPCFV
SPMSV
C-6
SPCaLV
SPCSV
SPVG
CMV

2.0 ELISA

2.1. *Ipomoea setosa*

3.0. Transmission of plant viruses

I.   Mechanical and by contact

II. (Grafting) ← (universal method for transmitting viruses) — **Sweetpotato**

III. Vectors

     Insects (aphids, whiteflies)

IV. Pollen and sexual seed

Wedge-grafting to Ipomoea setosa

Symptoms 1: Vein clearing
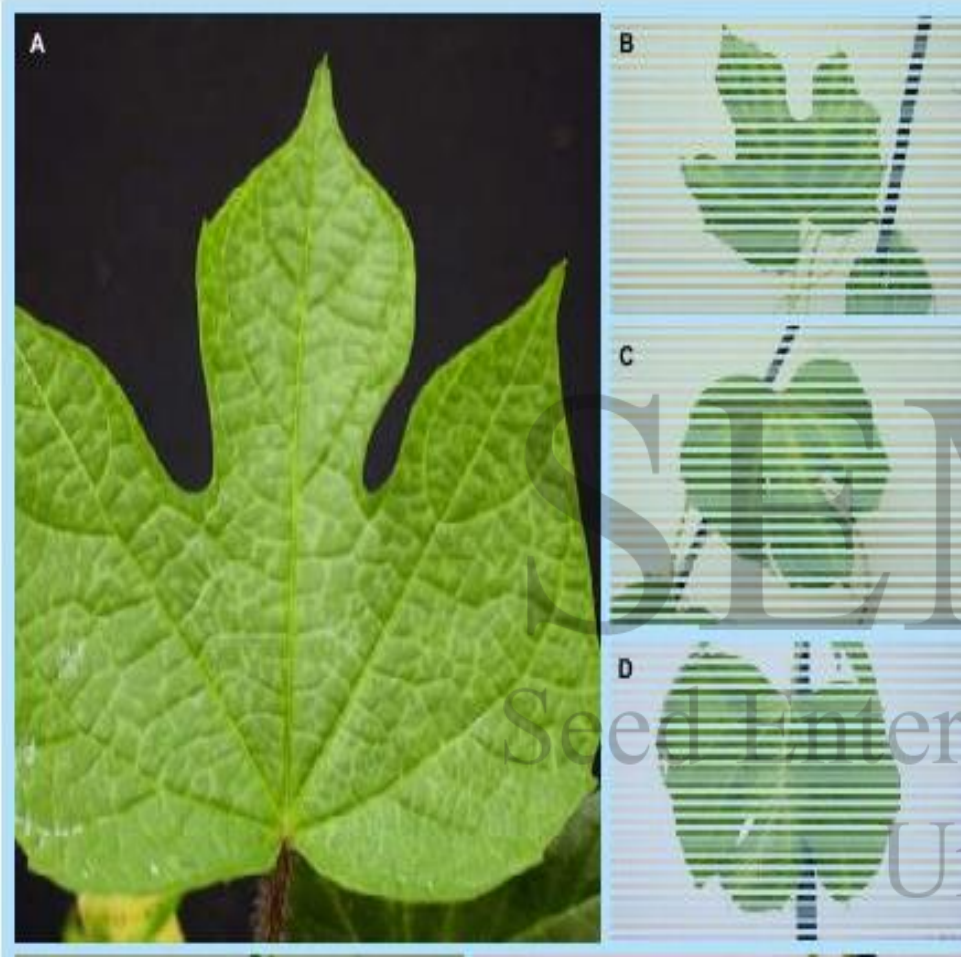
Symptoms 4:

a)Healthy   b)vein necrosis   b)chlorosis   c) vein clearing

Final Results of Virus Indexing

	Record	Other code	Accession number	Labcode	Collecting number	Accession name	SP symptoms	*I. setosa* symptoms	Serology	PCR	Results	Observations
38	35	1	CIP 401066	SG-1477	GTM 392	Camote Morado		Vc,Vn			REJECTED	
39	36	2	CIP 401448	SG-1524	DLP 2888	Morada					APROVED	
40	37	2	CIP 403380	SG-1558	INIVIT 9	Blanco Especial					APROVED	
41	38	2	CIP 402666	SG-1588	INIVIT1049			RU,D,IVC		Begomo	REJECTED	
42	39	5	CIP 402631	SG-1592	INIVIT1078			C,RU		Begomo	REJECTED	
43	40	5	CIP 402721	SG-1604	INIVIT1216			RU		Begomo	REJECTED	
44	41	6	CIP 401130	SG-1681	GTM 1108	Camote Morado		IVC		Begomo	REJECTED	
45	42	7	CIP 401375	SG-1688	SVG 27	Six Weeks		D,IVC		Begomo	REJECTED	
46	43	4	CIP 402619	SG-1702	INIVIT 217			RU		Begomo	REJECTED	
47	44	3	CIP 402655	SG-1717	INIVIT1038			D,Vc,Vn,Ln	SPCaLV		REJECTED	SPCaLV (PCR)
48	45	5	CIP 402696	SG-1731	INIVIT1084			C,IVC		Begomo	REJECTED	
49	46	1	CIP 403435	SG-1786	CESDA861607	86-1607					APROVED	

Control of viruses

1) **Always use planting materials cut from healthy-looking plants.**

- Make sure the whole field that you take your planting materials from is healthy, as this will reduce the likelihood of taking cuttings from plants that have just been infected but are not yet showing symptoms.

- Collect cuttings from young or mature crops (3-4 months old), as sweetpotato virus disease symptoms are harder to see in older plants.

2) **Remove and burn or feed to livestock any diseased plants as soon as they appear in young crops.**

- Roguing and is important to reduce the spread of the virus within your sweetpotato field. Iif a plant becomes infected it won't yield very much so you are improving your chances of harvesting a good yield if you remove infected plants as early as possible.

Control of viruses

3. Plant sweetpotato varieties that are resistant to the disease.

- Some varieties are rarely affected by virus disease. You can set up a replicated trial to test which varieties seem to be resistant to virus diseases.

4. Avoid planting new sweetpotato crops where you grew sweetpotato previous season.

- If there are roots or vines from old diseased plants surviving in the soil, they may sprout and produce diseased plants from which infection will easily spread to your new crop.

5) **Plant your new sweetpotato crop away from old sweetpotato crops.**

- This will make it harder for the aphids and whiteflies to reach your new crop and bring the virus disease from the old crop.

In vitro production of virus free plantlets
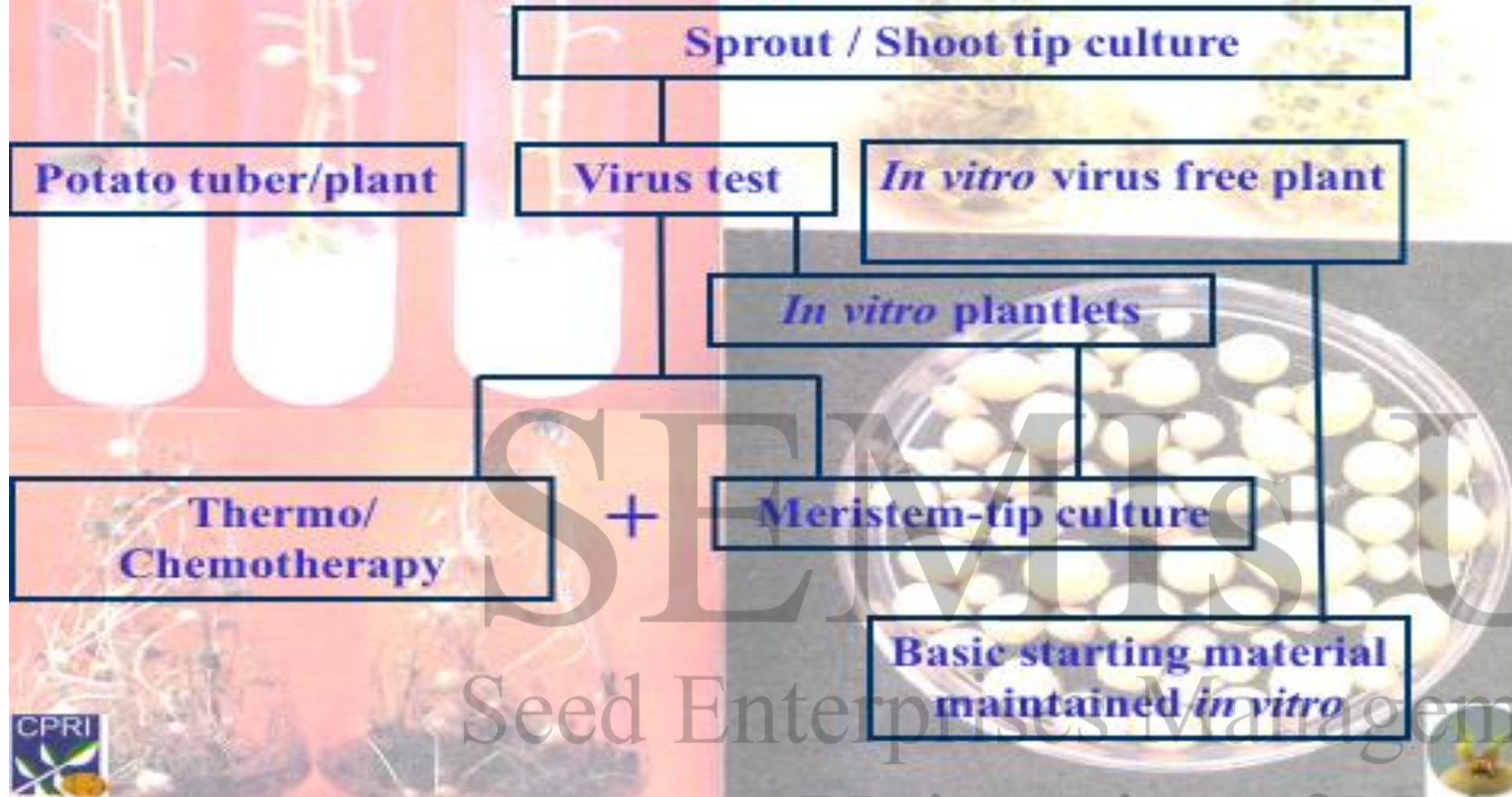
Tissue cultured based seed production system

Microplants → Microplants: 80-90% survival → 8-12 minitubers/plant → Generation-1 → Generation-2 → Breeder Seed

CLASSIFICATION PATHWAY

Kenya Plant Health Inspectorate Services (KEPHIS)

Micropropagation Laboratories

Pre-basic TC (ministem)

Class Pre-basic
Generation 1-4

Class Super Elite (SE)
Generation 1-3

**Class Elite (E)
Generation 1-3**

A
cannot be entered for further seed multiplication
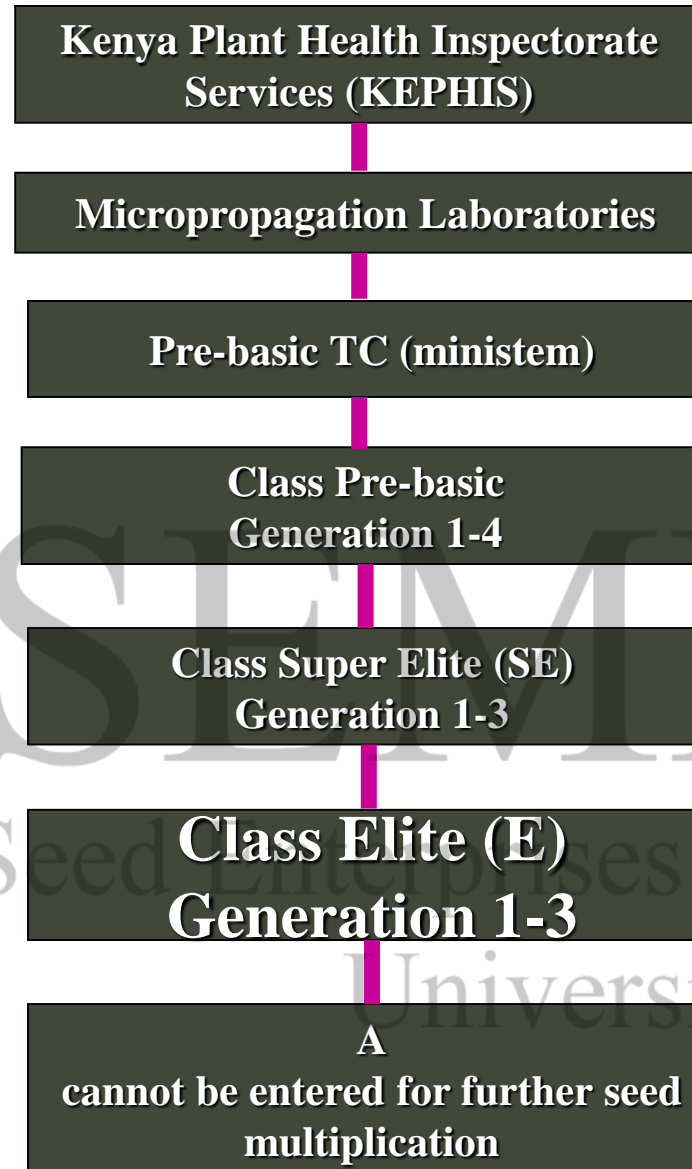
Generations in seed production and suggestion for virus detection

Greenhouse

- **G$_0$** — *In vitro* micropropagation
- **G$_1$** — Young plants

Test 100% of plants (composite samples by ELISA, indicator plants, NASH etc.

Field

- **G$_2$** — Test 25% of plants (composite samples by ELISA)
- **G$_3$** — Test 250 plants/ha by ELISA
- **G$_4$** — Test 250 plants/ 4ha by ELISA

Post-harvest test by symptomatology and ELISA

- **G$_5$**
- **G$_6$** — Symptomatology

Commercial production

G = Generation
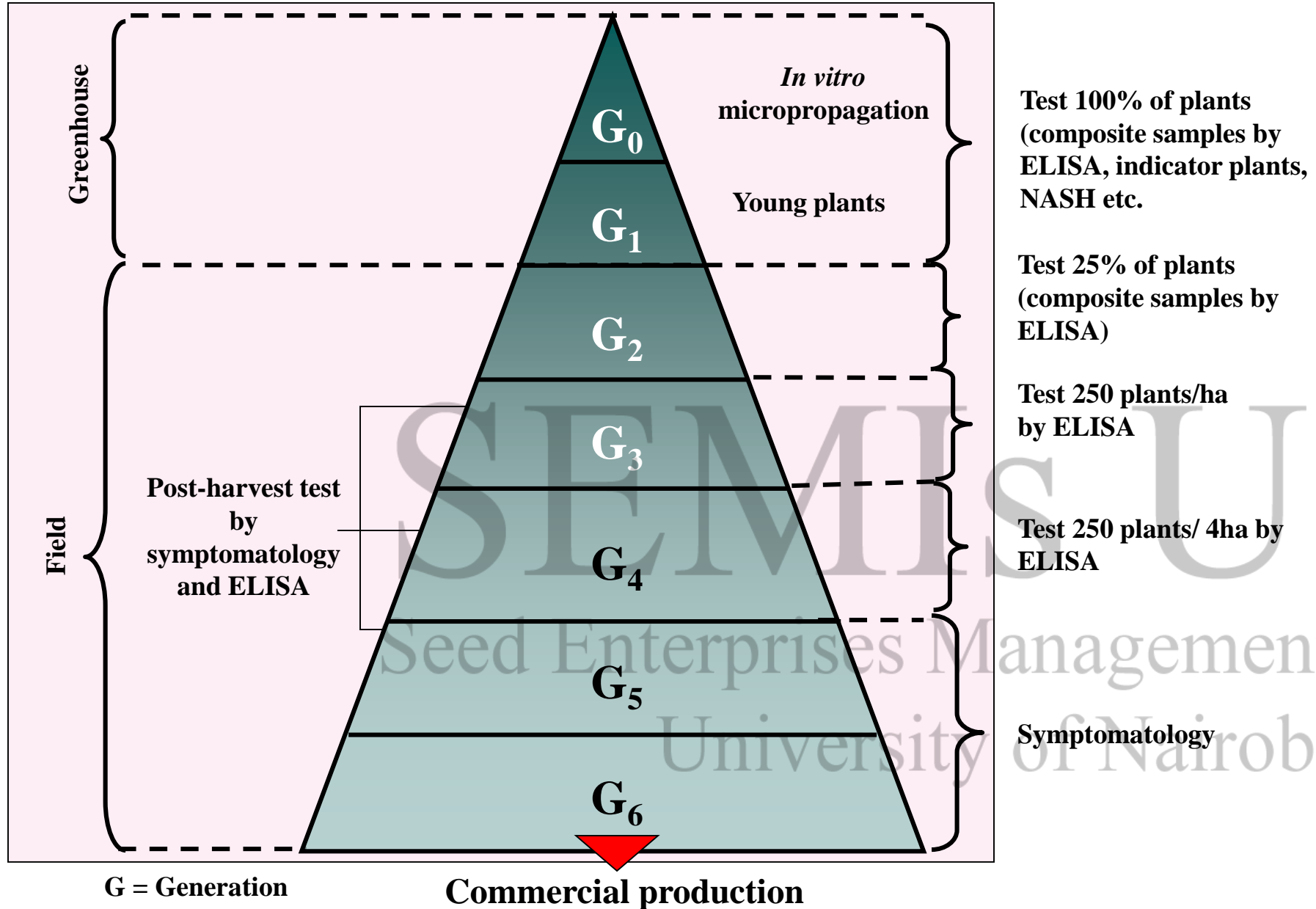
Seed Standards a case of the Netherlands (% tolerance)

Disease	Basic			Certified		
	S	SE	E	A	B	C
Field inspection						
Varietal impurity	0	0	0	0.01	0.05	0.1
Bacteria (Black leg)	0	0	0	0.03	0.1	0.1
Severe mosaic & PLRV	0.03	0.03	0.1	0.25	0.5	2.0
Mild mosaic	0.03	0.03	0.1	2.0	4.0	10.0
Total viruses	0.03	0.03	0.1	2.0	4.0	10.0
Verticillium wilt	2.0	2.0	3.0	4.0	8.0	-
Lot inspection						
Rhizoctonia (Blight)	10	10	25	25	25	25
Phytophthora	1 tuber per 50 Kg					
Common scab	Scab scale 2.5 (max. 1/8 of surface)					
External defects	4-12 tubers per 50 kg					

POTATO CERTIFIED SEED STANDARDS

Disease	Incidence			
	India	Holland[*]	Canada	UK
Mild mosaics	3	2	1	5
SM and PLRV	1	0.25	1	2
Brown rot	0.014	0	0	0
LB, Dry rot	1	NP	NP	NP
Wet rot	0	NP	NP	NP
Black scurf	5**	NP	NP	NP
Common scab	5**	NP	NP	NP
Total tuber Disease	5	-	-	-
Root knot nematode	Nil	NP	NP	Nil
Cyst nematode	Nil	NP	NP	Nil

NP : No limit prescribed

* : Brown rot prevalent since 1995

** : Tuber treatment compulsory