1. PEBRS (genetic pollution, biodiversity, escape genes, resistance to antibiotics, superweeds)
    . Resistance: gene flow
2. glyphosate is strongly adsorbed to the soil there is a negligible threat of residual effects on succeeding rotational crops. The number of herbicide applications in soybeans is estimated to have dropped by 12 percent for the period 1995-1999
3. Less contaminated crop for farmers
4. Shift in weeds present that must be controlled with other herbicides
    . payne and Oliver 2000: use normal post emergence herbicides to kill off resistant ones

ii.To farmers economy

1. dominant market for top agro companies as link their seed production to their chemicals. Monopoly might lead to loss of Biodiversity in traditional crops grown and also change farming practices so food supply can become vulnerable to changes in resistance in pests and pathogens.
2. fewer suppliers and seed saving illegal
3. More expensive seeds but should get return on non contaminated crop
4. Bayer bought monsanto and is now the biggest producer of seeds and agrochemicals 2019

b. Fungal resistance: Bt crops help reduce this

.to human health

i.to environment

3. Establish whether GM is better for environment and biodiversity. Experiments on:

. Efficiency and benefits of GM: how well they work in controlling:

.bacteria

i.virus

ii.fungi

iii.insect

iv.plant

1. Conventional plant breeding okay is good because lower seed cost and no import restriction but the seed cost is outweighed because of potential yield loss due to no resistance and import restrictions? Well cant get any through if it has fungal damage.

a. Agronomic benefits: Brookes 2014: 1996–2012 saw an increase of more than 370 million tons of food crops. One-seventh of the increased yield is attributed to GM crops in the U.S. To achieve an equal increase in yield as delivered by GM crops, it is estimated that an addition of more than 300 million acres of conventional crops would have been needed

b. Economic benefits: Brookes 2014: From 2006 to 2012, the global increase in farm income from GM food had reached $116 billion

4. When carrying out an ERA, the GMO Panel evaluates the overall impact of a GMO on the environment including :

. ecosystem services and endangered species

a. Is a GM plant, for example, more persistent and/or invasive than its conventional counterpart?

b. Does it have any adverse effects on organisms that are not meant to be affected by the GMO

the so-called non-target organisms

d. Impacts onxf biodiversity

i.main food produced in the UK: wheat (74%) (the most widely grown arable crop in the UK), barley, oats, potatoes, sugar beet, vegetables

ii.Main food produced in the USA: corn and grains, cotton, apples, strawberries, grapes

Themes

d. Aesthetics

.Altered flower colours of Torenia hybrida

i.Suppressed chalcone synthase and dihydroflavanol reductase to give white and blue colour

ii.Sterile flowers so wont go into food chain so no environmental concerns

iii.No limits to horticulture and pharmacological uses

5. Genetically Modifying plant Viruses

. Tobacco Mosaic Virus

.Particles are 18 x 300nm rods and genome is ss+ve sense RNA 6395 nucleotides long

i.Disassembly and replication

1. Virus enters cell through wound

. knows in host because of pH and ionic strength of cell solution

2. Uncoating: Ribosomes bind at 5' via a methylated G nucleotide and strip off coat proteins
3. Translation: Ribosomes bind to naked RNA and go to start codon, translate and fall off at stop codon = 126K protein
4. Stop codon leaky 10% of time so continue translating: 183K protein

. these are the replicase proteins: 126K and 183K

5. Subgenomic RNA production

. bind to 3' end of +ve sense RNA and make negative sense RNA.

a. This acts as template to make full length genomic +ve sense RNA and Replicases can skip first part of negative template and get shorter positive sense subgenomic RNA, ribosomes then bind and allows translation of MP and CP: 30K and 17.5K

ii.Replication

iii.Encapsidation: TMV Assembly

1. Coat proteins bind at origin of assembly and get encapsidation

iv.Cell to Cell Movement

1. Use movement protein to expand plasmodesmata so virus can get through, can slo pump naked RNA through

v.Long distance movement

1. reach phloem and this moves the virus all around plant

a. Preventing plant virus epidemics

.plant healthy seeds and tubers

1. needs a lot of testing so less possible in LEDCs

i.keep pl[illegible]

1. spray with insecticides as many insect vectors
2. but concern to environment

ii.Use resistant varieties

1. viruses mutate rapidly and overcome natural resistance

b. Cross protection against TMV

.mild strain inoculated against water control

i.mainly used in slow growing crops as takes long time to recover from virus

ii.Risks: can mutate and mild virus turn nasty

c. Targeting Transgenic resistance

.Coat protein mediated resistance

1. RNA for CP to dsRNA, add 35S promoter into A.tumefaciens and boom
2. Delayed symptom development , but could be infected with naked RNA

. Papaya ringspot virus: used CPMR was very effective

3. Believed because CPs were binding to naked RNA faster than ribosomes so didnt uncoat, but naked RNA ribosomes could bind

. less resistance with mutant CPs, more with better stability

a. Promoters driving expression in upper leaf epidermis, mesophyll or phloem. Only plants with accumulated CP in upper epidermis- site of infection showed resistance.

b. Very specific: resistance related to amino acid homology between virus and CP

i.TMV Movement Protein resistance

1. WT MP accelerated symptoms, remove amino acids from 3' terminus means it doesnt function

1. all occur naturally and to some extent in transgenic lines

6. Avoiding risks

. Synergism

.avoid sequences that trigger it like viral supressors of PTGS

a. transencapsidation

.use mutated coat proteins where amino acids involved in transmission are mutated

b. recombination

.include stop codons to prevent expression if recombine

e. Post transcriptional gene silencing

.Coat protein mediated resistance against potyviruses

1. aphid transmitted, with positive sense RNA, auto proteolytically cleaved to give 9 viral proteins
2. to express CP in transgenic plants need convert RNA to dscDNA and add start codon AUG instead of recognition site for protease

. resistant plants generated and had no RNA in cytoplasm and the more virus was thrown at the plant the more resistant it became

a. Transgene inside the nucleus: high levels of transcription inside nucleus but no RNA in the cytoplasm

3. POST TRANSCRIPTIONAL GENE SILENCING

. RNA made in nucleus and exits but when plant detects rNA levels over certain threshold targets it to be destroyed: particular sequence from virus is targeted to be destroyed so when add more virus it is destroyed as soon as it is exposed

a. Transgenic plants resistant to PVY have many PVY transgenes that seem highly transcribed but accumulate low levels of the transcript: resistance regulated by a cellular pathway that down regulates overexpressed mRNAs

b. Cytoplasmic based post transcriptional cellular surveillance system that targets specific RNAs

.aberrant RNA triggered this system that gives low steady state levels of the transgene RNA (like CP) when virus with same sequence enters plant it is targeted for elimination

i.Likely this with 54K, Movement protein as PTGS driving resistance

ii.Can take any part of virus, put in nucleus, screen for transgenic plants

1. use 35S promoter because strong and everywhere

c. RNA high level plant convert it to dsRNA and dicer enzyme cuts it into siRNA fragments 21bp long. Then binds to RNAse complex: RISC and destroys it

.when transform plants now put in piece of RNA

1. system doesnt need protein expression so no heteroencapsidation
2. no synergism as no protein
3. Homologous sequences from other viruses destroyed

4. Mubin 2019: PV2 protein encoded by papaya leaf curl virus renders it unsusceptible to Transcriptional gene silencing and PTGS

f. Virus induced gene silencing

.not used as mechanism to look at plant processes

i.have infectious clones to viruses so have TMV as DNA and insert genes: infects plant, reaches high levels and the insert is targeted and degraded

ii.Develop plants using this technology to silence genes

1. seeds from these plants are resistant: genetic imprinting

g. How do we get virus infections then?

.every virus has a gene that silences PTGS and potyviruses are the best at it

i.but this is a bad virus as it destroys its host, a good one keeps it alive

ii.Can get protein mediated resistance against viruses

1. mutated movement proteins give resistance and bind to plasmodesmata but most of time RNA is destroyed so how do you get the protein expressed in transgenic plants?

. expressing the bt gene and some have 35S promoter so bt gene is targeted and destroyed