Detection Methods for GM Foods in Korea

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1.Professor in Department of Food Sciences, Kyung Hee University, Korea (1996 – present)

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3. Committee member of GMO safety assessment in Feed of RDA (2003 – present)

4. Research Project Leader in GMO detection - Multiplex PCR ,Microarray, nano-technique

Background of GMO management in Korea

- 1. Increasing the mount of GMO development in the world
 - Global status of GMO Approval : 196 events in 24 crops (ISAAA, 2011)

2. Could not escape from GMO

- very low self-supply of crops in Korea (below 30%)
- Soybean ~5%, corn ~3%

3. Changes of consumer's awareness on Food safety

- increase the claim of food safety by NGOs against GMO
- Increase the need of well-being and organic foods

4. Manage to Unapproved GMO

- GM rice, GM papaya, GM flax , GM maize

National Framework of GMO Safety Assessment in Korea and Future Directions

Authorities for LMO/GMO Management



GM crops approved as Food by KFDA (76 events)

Crops	GM Events
Soybean (8)	RRS, A2704-12, MON89788, DP356043-5, DP305423-1, A5547-127, MON87701, DP305423-1xRRS
Maize (41)	MON810, TC1507, GA21, NK603, Bt11, T25, MON863, Bt176, DLL25, DBT418, DAS-59122-7, MON88017, Bt10, MIR604, MON89034, DP-098140-6, MIR162, 3272, MON863×NK603, MON863×MON810, MON810×NK603, MON810×GA21, 1507×NK603, MON810×MON863×NK603, DAS-59122-7×1507×NK603, 1507×DAS-59122-7, DAS-59122-7×NK603, Bt11×GA21, MON88017×MON810, Bt11×MIR604, Bt11×MIR604×GA21, MIR604×GA21, MON89034×MON88017, MON89034×NK603, MON89034×TC1507×MON88017×DAS-59122-7, 1507×DAS-59122-7×MON810×NK603, 1507×MON810×NK603, NK603×T25, MON89034×TC1507×NK603, Bt11×MIR162×MIR604×GA21, 1507×MIR604×NK603,
Cotton (15)	531, 757, 1445, 15985, 281/3006, LLcotton25, MON88913, GHB614,15985×1445, 531×1445, 15985×LLcotton25, MON15985×MON88913 281/3006×88913, 281/3006×1445, GHB614×LLcotton25
Canola (6)	GT73, Ms8/Rf3, T45, Ms1/Rf1, Ms1/Rf2, Topas 19/2
Potato (4)	SPBT02-05, RBBT06, Newleaf Y, Newleaf PLUS
Sugar beet (1)	H7-1
Alfalfa (1)	J101/J163
	(KFDA, 2012.4.)

Needs of GMO Detection method in Korea

For Approved GM food Labeling Systems

(Article 10 in Korean Food Sanitation Act)

- "Non GMO / Organic products / Korean products"
 - → Detect the existence of GMO (Qualitative)
- "Labeling-free products"
 - → Ascertain the Adventitious presence threshold level (≤ 3%) for authorized GMO

(IP Handling Certificates, Government Certificates, Quantitative)

For Mandatory Safety Assessment

(Article 4.6 in Korean Food Sanitation Act)

- Inspection of unapproved GMO (Qualitative)
- Prevent the import and distribution of unapproved GMO

□ For Risk Communication

- Post-market monitoring
- Right-to-Know for consumers

Multiplex PCR

> A simultaneous detection of various GMO in a single reaction, without loss of specificity

- Different events of GMO : soybean, maize, cotton, canola

Microarray

> An event-specific DNA microarray to identified GMO in Food





Development of a Multiplex PCR Method for Testing 6 GM Soybean Events



Detection of 8 events of GM Maize by Multiplex PCR



Fig. 1. Schematic diagram of eight events of GM maize.

Food Sci. Biotechnol. 15, 148 (2006)



Fig. 2. Multiplex PCR products amplified from 100% GM maize containing zein gene (endogenous gene). Lane 1 and 12: Marker; ØX174 DNA/HaeIII(TaKaRa, Japan), lane 2: Zein (endogenous gene, 99 bp), GA21(133 bp), T25(152 bp), TC1507 (172 bp), Mon810(193 bp), Mon863(223 bp), Event176(248 bp), Bt11 (265 bp) and NK603(314 bp), lane 3: Zein, lane 4: Zein and GA21, lane 5: Zein and T25, lane 6: Zein and TC1507, lane 7: Zein and Mon810, lane 8: Zein and Mon863, lane 9: Zein and Event176, lane 10: Zein and Bt11, lane 11: Zein and NK603.



Fig. 3. Multiplex PCR products amplified from various maize foods. Lane 1: Marker; ØX174 DNA/*Hae*III(TaKaRa, Japan), lane 2: Negative control, lane 3: Positive control, lane 4: Corn grits, lane 5: Cooking oil from corn, lane 6: Corn for feeding, lane 7: Canned corn, lane 8: Mixed vegetables, lane 9: Corn soup, lane 10: Corn chip, lane 11: Popcorn, lane 12: Cereal.

Multiplex PCR Detection of Four events of GM maize (Event3272, LY038, MIR162, and MON88017)

An event-specific multiplex PCR detection method for four events of GM maize was devised.

- Event 3272 : thermostable alpha amylase (amy797E) / mannose-6-phosphate isomerase (pmi)
- LY038 : dihydrodipicolinate synthase (cordapA)
- MIR162 : vegetative insecticidal protein (vip3A)
- MON88017 : delta-endotoxin (cry3Bb1) /cp4 epsps



Kim et al, (2009) J. Korean Soc. Appl. Biol. Chem. 52, 105

Multiplex PCR Detection of the MON1445, MON15985, MON88913, and LLcotton25 Varieties of GM Cotton

Event specific primers were used to distinguished 4 different GM cottons.



Fig. 1. Schematic diagram of 4 varieties of GM cotton.



Kim et al., (2008) Food Sci. Biotechnol. 17, 829

An event-specific DNA microarray to identify genetically modified organisms (GMOs) in processed foods.

We developed an event-specific DNA microarray system to identify 19 GMOs, including two GM soybeans (GTS-40-3-2 and A2704-12), thirteen GM maizes (Bt176, Bt11, MON810, MON863, NK603, GA21, T25, TC1507, Bt10, DAS59122-7, TC6275, MIR604, and LY038), three GM canolas (GT73, MS8 RF3, and T45), and one GM cotton (LLcotton25).

(A)

1	1	1	2	2	2	3	3	3	1	1	1
6	3	3	6	6	6	0	1	0	8	(8)	(8
٢	٢	٢	0	1	0	0	0	0	0	0	C
0	6	1	0	1	٥	6	6	(3	6	6	C
Ø	Ø	0	1	08	0	۲	19	0	0	20	0
1	1	0	0	0	0	3	0	3	0	3	C
٢	0	3	20	20	20	٢	2	0	20	2	2

No.	Targets	No.	Targets
1	blank	15	MON863
2	positive control	16	NK603
3	35S promoter	17	GA21
4	nos terminator	18	T25
5	loctin	19	TC1507
6	zSSlib	20	Bt10
7	FatA	21	DAS-59122-7
8	Acp1	22	MIR604
9	SPS	23	TC6275
10	GTS-40-3-2	24	LYO38
11	A2704-12	25	GT73
12	Bt176	26	MS8kRF3
13	Bt11	27	T45
14	MON810	28	LLcotton25



Kim et al., (2010) J. Agric. Food Chem. 58, 6018

Microarray Detection of GM Maize in Processed Foods

	GM maize events													
type of product	Bt176	Bt11	MON810	MON 863	NK603	GA21	T25	TC1507	Bt10	DAS-59122-7	MIR604	TC6275	LY038	no. of GM maize
nacho chips A	_	_	_	_	_	_	_	_	_	_	_	_	_	0
nacho chips B	_	_	_	_	_	_	_	_	_	_	_	_	_	0
nacho chips C	_	_	+	_	+	_	_	_	_	+	_	_	_	3
nacho chips D	_	_	-	-	_	-	_	_	_	_	_	_	_	0
nacho chips E	_	_	-	_	_	-	_	_	_	_	_	_	_	0
nacho chips F	_	_	+	_	+	_	_	+	_	+	_	_	_	4
nacho chips G	_	_	_	_	_	_	_	_	_	_	_	_	_	0
com chips	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn soup A	_	_	+	_	+	_	_	_	_	_	_	_	_	2
corn soup B	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn soup C	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn soup D	_	_	+	_	+	+	_	+	_	+	_	_	_	5
corn soup E	_	_	_	_	_	_	_	_	_	_	_	_	_	0
com soup F	_	_	_	_	_	_	_	_	_	_	_	_	_	0
canned corn soup	_	_	_	_	_	_	_	_	_	_	_	_	_	0
canned sweet com A	_	_	_	_	_	_	_	_	_	_	_	_	_	0
canned sweet corn B	_	_	_	_	_	_	_	_	_	_	_	_	_	0
canned sweet corn C	_	_	_	_	_	_	_	_	_	_	_	_	_	0
canned sweet corn D	_	_	_	_	_	_	_	_	_	_	_	_	_	0
popcom A	_	_	_	_	_	_	_	_	_	_	_	_	_	0
popcom B	_	_	_	_	_	_	_	_	_	_	_	_	_	0
popcom C	_	_	_	_	_	_	_	_	_	_	_	_	_	0
com flour A	_	_	_	_	+	_	_	_	_	_	_	_	_	1
com flour B	_	_	+	_	_	_	_	_	_	_	_	_	_	1
com flour C	_	_	_	_	_	_	_	_	_	_	_	_	_	0
com flour D	_	+	+	+	+	+	+	+	_	+	+	_	_	9
com flour E	_	<u> </u>	_	_	_	<u> </u>	<u> </u>	<u> </u>	_	_	_	_	_	0
corn bread A	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn bread B	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn bread C	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn bread mix A	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn bread mix B	_	_	_	_	_	_	_	_	_	_	_	_	_	0
corn bread mix C	_	_	+	_	+	_	_	+	_	+	_	_	_	4
com bread mix D	_	+	+	_	+	+	+	+	_	+	_	_	_	7
com starch A	_	_	+	_	_	_	_	_	_	_	_	_	_	1
com starch B	_	_	+	_	+	_	_	+	_	_	_	_	_	3
sausage containing com	_	_	_	_	_	_	_	_	_	_	_	_	_	ő
	type of product nacho chips A nacho chips B nacho chips C nacho chips D nacho chips F nacho chips F nacho chips G com soup A com soup A com soup D com soup C com soup C com soup C com soup F canned corn soup canned sweet com A canned sweet com B canned sweet com D popcom A popcom B popcom C com flour B com flour C com flour C com flour C com bread A com bread mix A com bread mix A com bread mix C com starch A com starch B sausage containing corn	type of productBt176nacho chips A-nacho chips B-nacho chips C-nacho chips D-nacho chips E-nacho chips F-nacho chips G-com chips G-com soup A-com soup C-com soup C-com soup C-com soup F-canned corn soup-canned sweet com A-canned sweet com D-canned sweet com D-popcom A-popcom C-com flour B-com flour C-com flour C-com flour C-com bread A-com bread Mix A-com bread mix A-com bread mix C-com bread mix C-com bread mix C-com starch A-com starch B-com	type of productBt176Bt111nacho chips Anacho chips Bnacho chips Cnacho chips Dnacho chips Enacho chips Fnacho chips Gcom chips Gcom soup Acom soup Ccom soup Ccom soup Ccom soup Fcanned sweet com Acanned sweet com Bcanned sweet com Cpopcom Apopcom Ccom flour Acom flour Bcom flour Ccom bread Mix Acom bread mix Bcom bread mix Ccom bread mix D+-com starch Acom starch Acom starch Acom starch Acom starch A <td>type of product Bt176 Bt11 MON810 nacho chips A - 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Microbead-assisted PDA sensor for the detection of genetically modified organisms

A simple and sensitive approach for the detection of marker protein, phosphinothricin acetyltransferase (PAT), from GM crops was developed based on the colorimetric transition of polydiacetylene (PDA) vesicles in combination with silica microbeads



Lim et al., (2011) Anal. Bioanal. Chem. 400, 777

Unauthorized GMOs in Korea

- GMOs that are approved in other countries but not in Korea or that have not been approved elsewhere in the world.
- The Korea threshold for Unauthorized GMOs is zero, which means that their presence is not allowed in food or feed.

- ✓ Flax
- ✓ Rice
- ✓ Papaya
- ✓ Tomato
- ✓ Carnation
- ✓ Rose





Simultaneous Detection of Unapproved GM Rice using Event-specific Primers



Conclusion

- Development of Detection methods
 - Multiple GM crops in Food
 - Unauthorized GMO
 - Stacked GMO
 - New approaches : Nanobiotechnology
- ✤ International Collaboration
 - Genetic Information: database
 - Standard materials
 - Interlaboratory test











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