plastic bag. The calibration curve for determination was adjusted for frozen temperature, since the film was less sensitive at cool temperature. After irradiation, samples were stored at −20 °C until analysis. The irradiated samples were analyzed within 1 month after irradiation. Nonirradiated control samples were also stored under the same conditions.

Extraction

A 5-g aliquot of sample and 5 g of diatomaceous earth particles as a drying agent were ground in a mortar (12 cm i.d.) with a pestle until the mixture became homogeneous to facilitate solvent penetration into the sample matrix. The mixture was transferred into a 50-mL polypropylene conical test tube and shaken vigorously for 1 min after the addition of 30 mL of *n*-hexane. The mixture was centrifuged (1,290×*g*, 10 min) with a Himac CR-GIII (Hitachi Koki, Tokyo, Japan). The supernatant was filtered with a filter paper (No. 5A, 125 mm, Advantec, Tokyo, Japan), and the precipitate was extracted with 20 mL of *n*-hexane again. These extracts were combined and evaporated. The remaining fat residue in the flask was heated at 70 °C for 30 min to remove the *n*-hexane completely before measuring the weight of fat, because ACBs were stable below 100 °C (Obana et al. 2006). The fat also could be a keeper of ACBs. Thus, the heating procedure would keep the ACBs in the fat without decomposition and vaporizing.

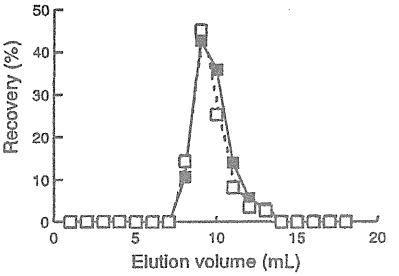

Fig. 3 Elution patterns of DCB and TCB with the handmade silica gel column. The *solid line with filled squares* indicates DCB; the *broken line with open squares* indicates TCB

Defatting

An aliquot of 0.2 g of fat residue was weighed into a glass tube with a screw cap and dissolved using 2.5 mL of acetone. After the addition of 0.5 mL acetonitrile, the sample was mixed using a vortex mixer for 1 min and then cooled at −20 °C for 30 min to precipitate the fat. The sample was centrifuged at 0 °C (1,290×*g*, 10 min), and the supernatant was collected in another glass tube to remove the precipitated fat. The solvent was completely removed with a nitrogen stream under warm conditions. The defatted sample was then weighed and dissolved in 2 mL *n*-hexane.

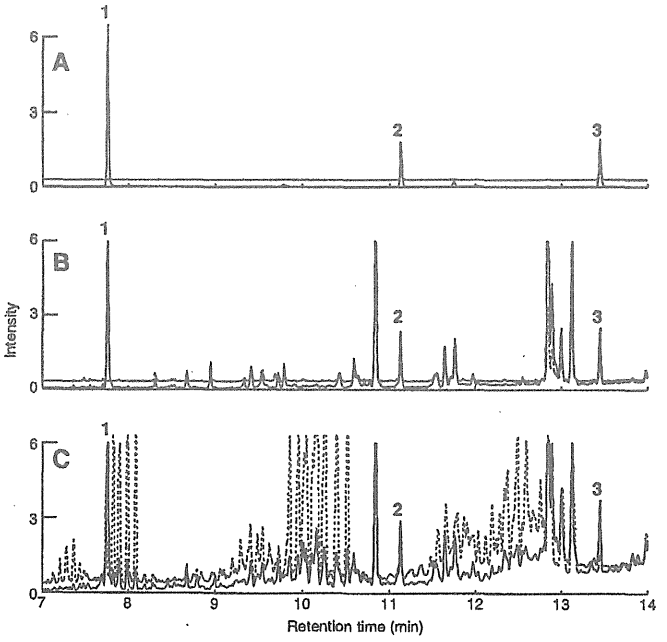

Fig. 2 GC-MS chromatograms of the test solution purified with the handmade silica gel column and the commercially available silica gel column: *A* DCB and TCB standard solution at 50 ng/mL, *B* the handmade silica gel column, and *C* the commercially available silica gel column. Peak labels: *1*, 2-cyclohexylcyclohexanone (I.S.); *2*, DCB; and *3*, TCB. The *solid line* indicates the monitoring ion (*m/z* 98); the *broken line* indicates the qualifier ion (*m/z* 112)

Table 1 Recovery rates of DCB and TCB in meat and processed foods

Sample	DCB		TCB	
	Mean[a] (%)	RSD[b] (%)	Mean[a] (%)	RSD[b] (%)
Beef	84	2	70	5
Pork	76	4	77	2
Parmesan cheese	67	12	73	5
Fried chicken	81	3	86	3
Hamburger	74	4	76	2
Gyoza	88	5	86	2
Gyudon	72	7	71	7

[a] Means of three experiments

[b] Relative standard deviation of three experiments

Cleanup

The defatted sample was passed through the handmade silica column prepared as described above. For a defatted sample weighing over 0.06 g, the sample load was divided between two silica gel columns. Ten milliliters of *n*-hexane and 5 mL of 2 % diethyl ether in *n*-hexane were eluted and discarded. The DCB and TCB fraction was eluted with 10 mL of 2 % diethyl ether in *n*-hexane and then dried with a nitrogen stream under warm conditions. The DCB and TCB fraction was reconstituted with 0.2 mL *n*-hexane containing 100 ng/mL of 2-cyclohexylcyclohexanone as I.S.

GC-MS Determination

The GC-MS used was QP2010 Ultra (Shimadzu, Kyoto, Japan). The GC conditions were as follows: column, DB-5MS (Agilent, CA, USA), i.d. 30 m×0.25 mm, thickness 0.25 μm; column temperature program, 60 °C (1 min), 60–160 °C at 20 °C/min, 160–250 °C at 8 °C/min, 250–300 °C at 25 °C/min, 300 °C (5 min); carrier gas, He; injection temperature, 250 °C; injection volume, 1 μL; and injection mode, pulsed splitless. The MS conditions were as follows: ionization mode, electron ionization; ion detection, scan (*m/z* 95–115) and selected ion monitoring (*m/z* 98, 112); ionization voltage, 70 eV; ion source temperature, 200 °C; and transfer line temperature, 250 °C.

Calibration Curves for DCB and TCB Measurements

To compensate for the effects of any sample matrix remaining in the test solution on GC-MS analysis, DCB and TCB were quantified using an internal standard method. The relative

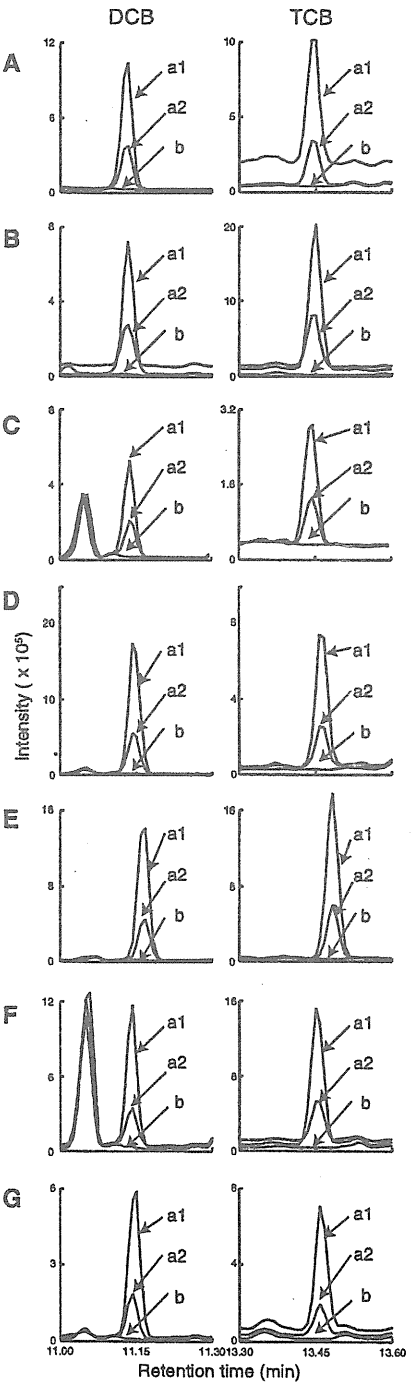

Fig. 4 SIM chromatograms of DCB and TCB in irradiated food samples monitored at *m/z* 98: *A* beef, *B* pork, *C* Parmesan cheese, *D* fried chicken, *E* hamburger, *F* *gyoza*, and *G* *gyudon* (*a1*, irradiated at 2.6 kGy; *a2*, irradiated at 1.0 kGy; and *b*, nonirradiated)

peak area of DCB and TCB against the internal standard at 100 ng/mL was calculated for calibration.

Results and Discussion

Method Development for DCB and TCB

A typical method for the determination of DCB and TCB consists of the following three steps: (1) a fat extraction step, (2) a cleanup step including defatting, and (3) a GC-MS analysis step. Although the EN1785 (European Committee for Standardization 2003) method aims to extract all the fat in the sample with a Soxhlet extractor, the fat required for DCB and TCB determination would be an aliquot of that; only 0.2 g of fat is used. The time for extraction of fat with a Soxhlet extractor would be approximately 6 h. Furthermore, this process consumes solvent in large quantities; thus, direct extraction with solvent would be suitable for the development of a rapid and simple method for the determination of DCB and TCB. In this study, direct extraction with solvent was adopted as previously described (Hijaz et al. 2010; Tewfik 2008).

The extracted fat was processed with defatting and silica gel column cleanup before GC-MS analysis. Cleanup is the key step for developing a reliable and robust method for matrix-rich samples such as fatty foods. In EN1785 (European Committee for Standardization 2003), after fat extraction with Soxhlet apparatus, the cleanup depends only on a Florisil column. The cleanup step in EN1785 (European Committee for Standardization 2003) could be unsatisfactory for routine analysis because of the large-sized column and the large quantity of solvents used. To obtain a clear chromatogram for DCB and TCB, a selective extraction of DCB and TCB using an SFE system or further column cleanup would be required (Stewart et al. 2001). In the present study, freezing out the fat in the solvent and a silica gel column were used for the cleanup process. In both these processes, special apparatus, such as SFE or ASE, were not used.

The proposed method could provide a versatile data expression, comparing with EN1785 (European Committee for Standardization 2003) and the previous method (Tewfik et al. 1999; Horvatovich et al. 2000; Stewart et al. 2001; Gadgil et al. 2002; Hijaz et al. 2010; Tewfik 2008). The DCB and TCB concentrations could be expressed on two bases, the fat basis or as sample weight, which enable comparisons with results from previous reports that use various ways to express the DCB and TCB concentrations in samples. The EN1785 method expressed results on the fat basis (European Committee for Standardization 2003) while SFE and the direct solvent extraction method expressed concentrations on the sample weight basis (Tewfik et al. 1999; Horvatovich et al. 2000; Stewart et al. 2001; Gadgil et al. 2002; Hijaz et al. 2010; Tewfik 2008).

The Fat Extraction Step

The efficiency of fat extraction was compared between this method and a method using a Soxhlet extractor with a 5-g sample of beef in duplicate. The average of fat obtained by the two methods was 1.34 and 1.24 g, respectively, and there would be no significant differences between these methods in the efficiency of fat extraction. Furthermore, the times for extraction from the sample were 90 min and 1 day, respectively. The fat extraction step could be replaced by the direct solvent method.

The Cleanup Step

The fat extract was defatted before passing through a silica gel column to avoid overloading. To remove almost all the fat

Table 2 Concentration of radiation-induced DCB and TCB in meat and processed foods

Sample	DCB						TCB					
	0 kGy		1.0 kGy		2.6 kGy		0 kGy		1.0 kGy		2.6 kGy	
	Mean[a]	RSD[b]	Mean[a]	RSD[b]	Mean[a]	RSD[b]	Mean[a]	RSD[b]	Mean[a]	RSD[b]	Mean[a]	RSD[b]
Beef	ND	–	86	15	243	9	ND	–	65	8	185	7
Pork	ND	–	50	9	138	1	ND	–	139	8	361	6
Parmesan cheese	ND	–	72	8	168	6	ND	–	36	4	90	6
Fried chicken	ND	–	127	5	385	2	ND	–	48	2	155	2
Hamburger	ND	–	110	3	314	4	ND	–	118	3	335	4
Gyoza	ND	–	72	5	193	2	ND	–	92	7	272	1
Gyudon	ND	–	69	5	236	9	ND	–	61	6	243	8

ND not detected

[a] Means of three experiments (in nanograms per gram lipid)

[b] Relative standard deviation of three experiments (in percent)

from the fat extract, acetonitrile, a fat-insoluble solvent, was added to the fat extract dissolved in acetone and then cooled at −20 °C. After cooling, the insoluble fat precipitated and was easily removed by centrifugation. The optimal ratio of acetone to acetonitrile was 5:1 (2.5 and 0.5 mL). If the ratio was less than 5, the recovery of DCB and TCB declined; for a ratio more than 5, the fat remaining increased (data not shown).

We found that when using commercially available silica gel columns, unknown contaminants were eluted from the column to the DCB and TCB fraction. These contaminants overlapped both the SIM (*m/z* 98 and 112) of DCB and TCB in GC-MS analysis (Fig. 2(C)). Even after proper conditioning before use, contaminants were detected from all the available silica gel columns we tested. To obtain further information of the contaminants, the extracts of one of commercially available silica gel column parts (cylinder unit, flit, and silica gel) with the eluate were analyzed in the same GC-MS conditions. The contaminants were mainly detected from the extract of the cylinder unit (data not shown). The peaks of contaminants consecutively appeared on chromatograms as shown in Fig. 2(C). The cylinder unit was made of polypropylene resin. Thus, the contaminants could be a mixture of paraffin- and/or olefin-like compounds reached from the cylinder units. The interference from these contaminants was successfully avoided by using our handmade silica gel column in which the usage of resin was minimal (Fig. 2(B)).

Figure 3 shows the typical elution profile of the silica gel column loaded with a defatted extract (0.04 g) obtained from 0.2 g beef fat containing 80 μg each of DCB and TCB. After washing with 10 mL of *n*-hexane, 20 mL of 2 % diethyl ether in *n*-hexane was eluted and fractionated with every 1 mL. DCB and TCB were eluted into 5–15-mL fractions. The recovery rates of DCB and TCB from the column were 112 and 101 %, respectively.

Recovery Tests

In the recovery tests, fat extracted from nonirradiated foods was used as a blank sample. The blank samples (each 200-mg portion), spiked with DCB and TCB at 50 ng/g each, were used as positive samples. Recovery tests were conducted in triplicate for each food sample (Table 1). The mean values of recovery of DCB and TCB were 70–88 % with less than a 10 % relative standard deviation (RSD), except for the Parmesan cheese (DCB 67 %). This method would be applicable for the determination of DCB and TCB in almost all irradiated foods.

Analysis of Irradiated Food

The DCB and TCB levels in the food samples (beef, pork, Parmesan cheese, fried chicken, hamburger, *gyoza*, and *gyudon*) irradiated at two dose levels, 1.0 and 2.6 kGy, were determined by our method in triplicate. Figure 4 illustrates typical chromatograms of DCB and TCB irradiated at two dose levels and of nonirradiated samples. Both the chromatograms at *m/z* 98 and 112 were clear enough to identify and quantify both DCB and TCB (data not shown). Significant interference peaks could not be found beneath those for DCB and TCB in the negative control samples. DCB and TCB were detected in all irradiated samples at levels depending on the dose, but in none of the nonirradiated control samples (Table 2). Both DCB and TCB levels in these experiments also resembled those observed in our former reports (Obana et al. 2005, 2006), where the concentrations of DCB and TCB were determined in meat irradiated under similar conditions.

In conclusion, this method is simple, is rapid, and requires no special system for the preparation of the test solutions for GC-MS analysis. The time to prepare test solutions for GC-MS analysis from three samples was approximately 8 h by this method. This method would be a potential candidate for a routine analysis method for determining DCB and TCB levels in lipid-rich foods.

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Conflict of Interest Yoko Kitagawa declares that she has no conflict of interest. Masahiro Okihashi declares that he has no conflict of interest. Satoshi Takatori declares that he has no conflict of interest. Keiji Kajimura declares that he has no conflict of interest. Hirotaka Obana declares that he has no conflict of interest. Furuta Masakazu declares that he has no conflict of interest. Toshimasa Nishiyama declares that he has no conflict of interest. This article does not contain any studies with human or animal subjects.

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食品中放射性物質の安全確保対策

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平成15年の食品安全委員会による食品安全モニター・アンケート調査「食の安全性に関する意識調査」において、食品の安全性の観点からより不安を感じているものの１番は農薬（67.7%）、以下、輸入食品（66.4%）、添加物（64.4%）、汚染物質（60.7%；ダイオキシン、環境ホルモン、重金属など）でした。食中毒発生件数原因の８割、食中毒患者数で９割以上を占める微生物、ウィルスについての不安は、それぞれ７番（46.8%）、11番（34.3%）でした。今、同様のアンケート調査を行ったならば、生食肉による食中毒事件への関心から微生物への不安も高くなるでしょうが、他を圧倒して関心が高いのは放射性物質汚染でしょう。

放射線は見えず、音もせず、触れず、味も臭いもない、人の五感で探知できません。専用の測定機器でなければ正確に検出できない性質ゆえに、放射線に対する警戒は高まらざるをえないのではないでしょうか。放射線を発する物質を放射性物質、放射線の強度を放射能といいます。放射能は１秒間に崩壊する原子核の数；Bq（ベクレル）で表されます。放射線は電離作用（主に分子中の電子を離脱させることによります）によって、人体に障害を引き起こします。特に分裂期の細胞は、通常であれば細胞核、染色体、二重らせん構造により三重に守られている遺伝子がフリーの状態にあるためダメージを受けやすく、健全な細胞分裂が阻害されます。したがって、細胞増殖の活発な胎児、乳幼児への影響が大きいと考えられる一方、放射線は無秩序に増殖するガン細胞などの腫瘍の治療や、増殖の速い細菌類の殺菌、滅菌に用いられています。人の放射線被曝の指標はSv（シーベルト）で表され、日本人の医療目的等を除く自然放射線被曝量は平均で年間1.5mSv、世界平均では年間2.4mSvとされています。

食品の放射性物質汚染についての基準としては、1986年チェルノブイリ原子力発電所事故を受けて、輸入食品中の放射能暫定限度、セシウム134とセシウム137の和として370Bq/kgがありました。３月の放射性物質放出事故を受けて、食品中放射性物質の暫定規制値（表１）が定められましたが、規格基準、規制のあり方等についての初期の説明不足は否めず、暫定規制値で健康が確保できるのか、流通している食品は安全なのかなどの不安は払拭されませんでした。10月に「食品中に含まれる放射性物質の食品健康影響評価」が示され、食の健全性を守るための規格基準設定に向けての取組が始まりました。

食品中に含まれる放射性物質の食品健康影響評価

厚生労働省は３月17日、「放射能汚染された食品の取り扱いについて（食安発0317第３号）」と題して食品中放射性物質の暫定規制値およびその検査方法を示しました。暫定規制

表１　飲食物摂取制限に関する指標（2011年３月17日厚生労働省）

核種	原子力施設等の防災対策に係る指針における摂取制限に関する指標値（Bq/kg）	
放射性ヨウ素（混合核種の代表核種：^{131}I）	飲料水、牛乳・乳製品 注)	300
	野菜類（根菜、芋類を除く）、魚介類（４月５日追加）	2,000
放射性セシウム	飲料水、牛乳・乳製品	200
	野菜類、穀類、肉・卵・魚・その他	500
ウラン	乳幼児用食品、飲料水、牛乳・乳製品	20
	野菜類、穀類、肉・卵・魚・その他	100
プルトニウム及び超ウラン元素のアルファ核種*	乳幼児用食品、飲料水、牛乳・乳製品	1
	野菜類、穀類、肉・卵・魚・その他	10

注) 100 Bq/kgを超える牛乳・乳製品は、乳児用調製粉乳及び直接飲用に供する乳に使用しないよう指導すること。

* ^{238}Pu, ^{239}Pu, ^{240}Pu, ^{242}Pu, ^{241}Am, ^{242}Cm, ^{243}Cm, ^{244}Cm 放射能濃度の合計

値は、平成10年に示された原子力安全委員会による「飲食物摂取制限に関する指標について」に基づいており、行政機関が公衆の放射線防御のために対策をとるべきレベル（介入線量レベル）として、当該食品の出荷、流通、摂食を制限することができる指標値であり、毒性試験等の結果をふまえた科学的評価に基づく環境汚染物質、農薬等の残留基準値（人が生涯にわたり継続して摂取しても健康に影響がない食品中残留量）とは性質が異なります。介入線量レベルは、ICRP（International Commission on Radiological Protection：国際放射線防護委員会）の勧告を基にしており、年間実効線量５mSvを限度としています。

内閣府食品安全委員会は、３月29日付「放射性物質に関する緊急とりまとめ」により暫定規制値をおおむね追認した後、７月26日に「食品中に含まれる放射性物質の食品健康影響評価（案）」を策定し、パブリックコメントを経て10月29日に最終的な評価書を示しました。その概要は、ウランの耐容一日摂取量（人が生涯にわたり継続して摂取しても健康に悪影響を及ぼすおそれがないと推定される１日当たりの摂取量）を0.2μg/kg体重/日（体重50kgであれば１日当たり10μg）とするほか、放射線の健康影響については悪影響が見いだされるのは、通常の一般生活において受ける放射線量を除いた生涯における累積線量として、おおよそ100mSv以上としています。

評価を受けて厚生労働省は10月28日、「食品中の放射性物質の規制値の設定について」にて、許容できる線量を年間１mSvに引き下げることを基本として、規制値設定のための検討を進めていく方針を明らかにしました。年間線量１mSvとは食品から経口摂取した場合、ヨウ素131、セシウム134、セシウム137として各単独で45,500Bq、52,600Bq、76,900Bq相当になります。１日あたりに換算するとヨウ素131、セシウム134、セシウム137として各単独で125Bq、144Bq、211Bqです。事故後８か月目のセシウム134、セシウム137の存在比を８：10として計算すると、放射性セシウムを１日あたり181Bq経口摂取すると年間線量１mSvとなります。現行の暫定規制値は年間線量５mSvを基に策定されておりますが、年間線量１mSvに対応するための規制値は単純計算で現行の１/５になります。

食のリスクマネージメント

話が前後しますが、日本における食のリスクマネージメントについて紹介します。

農薬、添加物、汚染物質といった化学物質をはじめとして、微生物、ウィルス、遺伝子組換え食品、新開発食品、その他食品関連分野において健康危害要因となりうる項目については、内閣府食品安全委員会がリスク評価を行います。具体的に農薬を例として説明します（図１）。国内で使用できる農薬は農薬取締法に基づき登録されていなければなりません。①登録の窓口は農林水産省です。②農薬の登録申請を受けた農林水産省は、食品衛生法に基づく残留基準の設定を厚生労働省へ、また、農薬取締法に基づく環境影響評価を環境省へ要請します。③厚生労働省は残留基準設定のための健康影響評価を食品安全委員会へ要請します。食品安全委員会は、規制や指導等のリスク管理を行う関係行政機関から独立して、科学的知見に基づき客観的かつ中立公正にリスク評価を行うことを使命としており、当該農薬について各種の毒性試験結果等から科学的に健康への影響についてリスク評価します。過去の評価例には、人の健康に影響をおよぼさない摂取許容量を設定する場合、影響が認められないとして評価の必要なしとする場合、毒性が高いため摂取許容量を設定できないとする場合、科学的評価のためのデータが足りないとして評価を保留する場合などがあります。④食品安全委員会による健康影響評価を受けて、厚生労働省は残留基準を設定します。通常は日本人の摂食状況を考慮して、摂取許容量を超えないように各種食品に対して残留基準を設定します。⑤厚生労働省による残留基準設定および環境省による環境影響評価を受けて、農林水産省は当該農薬の使用基準等を定めて、⑥登録します。

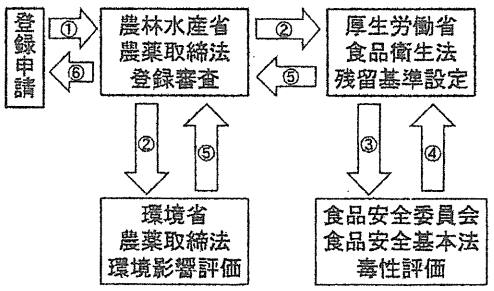


図１　食品中残留農薬のリスクマネージメント

以上、各行政機関の役割を示しましたが、各行政機関は主要ステップごとに、パブリックコメント、説明会等を実施して、施策案を消費者、事業者、民間団体等に周知するとともに、意見を求めて施策に反映させていく体制をとっています。施策実施後の監視、指導も各行政機関の役目です。

食品中の放射性物質、特に原子力関係施設の災害、事故により放出された放射性ヨウ素、セシウムについての健康影響に関するデータは、当該事故時のものを参考にするしかありません。医療目的で使用される放射性物質については、健康影響評価のためのデータを必要に応じて集めることができるでしょうし、評価されて安全性が確保されるまで使用されないでしょう。評価を待つ間にも食事は摂らなければなりませんので、食品について同様の対応をとることは困難です。輸入食品に頼るにも限界がありますし、平成21年度までは370Bqを超える輸入食品が検疫所において確認されていたことから、輸入食品の全てが安全なわけではありません。輸入食品に依存することにより、国内の食品生産、製造者へのダメージ、それに伴う国益全体への影響も心配です。

食品中の放射性物質検査

食品安全委員会による「食品中に含まれる放射性物質の食品健康影響評価」を受けて、厚生労働省は現行の暫定規制値の根拠である年間線量5 mSvを将来的に1 mSv以下に引き下げ、生涯累積線量が100mSv以下となるように規制値を設定するでしょう。規制値が引き下げられると、食品中の放射性物質検査の正確さがさらに重要になります。現行の暫定規制値は数百Bqレベルですので、数十Bqまで計ることができれば規制値に対する適否は判定できますが、規制値が数十Bqになれば数Bqまでの正確さが要求されます。また、測定値が正確であるほど、放射性物質の摂取状況、被曝状況をより厳密に推定できます。

食品中の放射性物質検査方法は、厚生労働省より「緊急時における食品の放射能測定マニュアル」(平成14年厚生労働省策定、平成23年3月17日厚生労働省通知) が示されています。その内容は、1.ヨウ化ナトリウムシンチレーションサーベイメータによる放射性ヨウ素の測定法、2.ゲルマニウム半導体検出器を用いたガンマ線スペクトロメトリーによる核種分析法、3.ウラン分析法及びプルトニウムの迅速分析法、4.放射性ストロンチウム分析法からなります。放射性物質放出事故直後は、放射性ヨウ素、セシウムによる広範囲の汚染が認められましたが、半減期の短い放射性ヨウ素は半年で元の5百万分の1以下になっており、現在検出されるのは放射性セシウムがほとんどです。ウラン、プルトニウム、ストロンチウムについては放射性セシウムに比べて問題になっておりませんが、放射性ヨウ素、セシウムの測定が可能なヨウ化ナトリウムシンチレーションスペクトロメーター (NaI-γ線測定装置)、ゲルマニウム半導体ガンマ線スペクトロメーター (Ge-γ線測定装置) が事故直後よりかなり普及して検査実数が増えたのに比べて、ウラン、プルトニウム、ストロンチウムを検査可能な試験検査機関がいまだに少ないため、実態を把握しきれていないのではないかとの指摘もあります。ウラン、プルトニウム、ストロンチウムの検査には、標準品として放射性物質を用いるので、放射性物質取扱資格、専用の施設、設備、機器、管理体制が必要となるため対応可能な機関が少ないのです。

Ge-γ線測定装置によるガンマ線分析の例を図2に示しました。検体は放射能既知の核種 (カドミウム109、セリウム139、コバルト

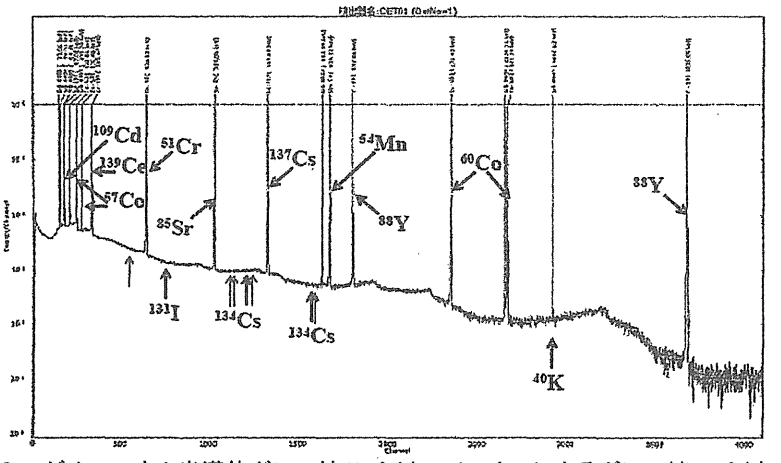


図2　ゲルマニウム半導体ガンマ線スペクトロメーターによるガンマ線スペクトル

57、クロム51、ストロンチウム85、セシウム137、マンガン54、イットリウム88、コバルト60）が充てんされたもので、ガンマ線分析における放射性物質の定性、定量のための標準線源として使用します。定性とは、放射性物質の種類を分別確認することです。放射性物質から放射されるガンマ線は、各放射性物質ごとに固有のエネルギーを持っています。図2の横軸はガンマ線のエネルギーを示しており、図の左に現れるエネルギーの低いカドミウム109（^{109}Cd）からエネルギーの高いイットリウム88（^{88}Y）まで、それぞれの放射性物質は決まった位置に現れます。セシウム134（^{134}Cs）などのように、複数のエネルギー順位を持つ放射性物質もあります。横軸のどの位置にあるか、すなわちガンマ線のエネルギーを調べることにより放射性物質の種類がわかります。一方、図2の縦軸は、計測されたガンマ線の量を表します。計測されたガンマ線の量から、放射性物質の放射能がわかります。これを定量といい、食品中の放射性物質の量が数値（Bq/kg）として求められます。

日本食品衛生協会においても、Ge-γ線測定装置による食品等の放射性ヨウ素、セシウムの検査を受託しておりますが、検査依頼者からの問い合わせが多いのは、検出限界と定量下限についてです。検出限界とは分析対象とする放射性物質の有無を判定できる最小値（Bq/kg）のことであり、定量下限とは定量した数値（定量値）の信頼性を確保できる最小値（Bq/kg）のことです。当協会では放射性物質の有無の判定および定量値ともに99％以上の信頼性を確保できる体制で検査しております。例えば、検出限界5 Bq/kg、定量下限50Bq/kgの場合には、検査結果が5 Bq/kgを超えるとき分析対象とする放射性物質が存在するという判定は99％以上の確率で正しく、検査結果が50Bq/kgを超えるとき定量値の信頼性は99％以上です。

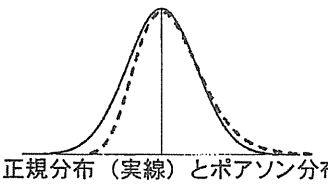


図3　正規分布（実線）とポアソン分布（破線）

Ge-γ線測定装置による検出限界の求め方の代表例はCooper法です。分析対象とする放射性物質のエネルギー位置におけるガンマ線量が、その前後の位置のガンマ線量の標準偏差の3倍以上認められた時を検出とし、標準偏差の3倍を検出限界値とします。理化学的分析分野においては、検出限界を標準偏差の3倍とすると、定量下限は標準偏差の10倍とすることが多いのですが、これは分析結果の分布が正規分布であることを前提としています。ところが放射線の分析結果の分布はポアソン分布であることが知られています。図3に示した通り、正規分布は平均値を中心に左右対称ですが、ポアソン分布は平均値より大きい側に歪んだ形になります。これは定量値が真の値よりも大きくなる可能性をはらんでいることになります。ただし、ポアソン分布はガンマ線のカウント数が1,000以上の場合、ほぼ正規分布とみなすことができるため、信頼性等の統計学的評価が可能となります。検体の量が多いほど、また測定時間が長いほど、ガンマ線のカウント数が多くなり、検出限界および定量下限を引き下げることができます。

検出限界、定量下限についての考慮のほかに検査の信頼性を確保するためには、測定装置の管理、検体の調製、測定結果の確認について熟知している必要があります。

測定装置の管理では、前に述べたような放射性物質の種類および放射能が明確な標準線源を、測定に用いる容器の種類毎に保有して、放射性物質の同定、放射能の強度を正確に測定するための装置の調整を随時行う必要があります。高濃度汚染検体の測定後には、ガンマ線スペクトル（図2）の横軸、縦軸にズレ

が生じることがあるとともに、装置自体が放射性物質に汚染される可能性についても配慮する必要があります。

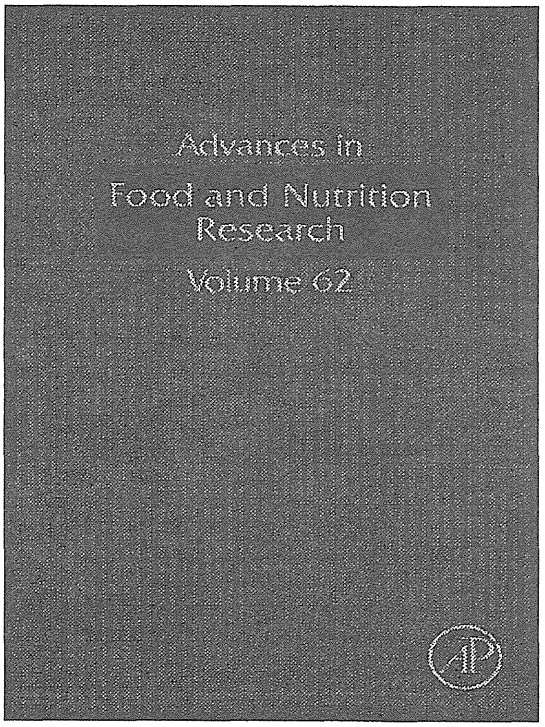
検体の調製では、検体の完全な均一化および空隙が最小限になるように測定用容器に充てんする必要があります。検出される放射能は、測定装置の検出部（ゲルマニウム半導体）と放射線源（放射性物質）の距離の二乗に反比例します。放射線源と検出部の距離が1cmのときに検出される放射能に比べて、距離が2cmの時の検出される放射能は4分の1、距離が10cmの時は100分の1になります。放射性物質が検体中に均一に存在するようにしなければ、正確な定量値は求められません。例えば、肉類における放射性セシウムの分布濃度は、筋肉部位で脂肪部位よりも2倍以上高いため、測定の際に測定装置の検出部の近くに脂肪部位が置かれた場合よりも、筋肉部位を置いた方が定量値は高くなります。同じ理由から、検体を測定用容器に充てんする際には、空隙を極力減らすようにしなければなりません。充てんされた検体中に生じた空隙には放射性物質は存在しませんので、放射性物質の偏在の原因になります。

測定結果の確認では、検出された放射性物質、定量値、検出限界等を再確認します。定期的に実施する測定装置の調整、汚染状況の確認の他にも、食品のほとんどに含有されているカリウム40を指標として、個々の測定が正しく行われているか確認することができます。分析対象とする放射性物質を間違いなく測定しているか、他の放射性物質を間違えて計っていないか、さらに必要とされる検出限界を満たしているか、再現性に問題はないかを確認した上で、定量値の正当性を判断します。

厚生労働省から示された、「食品中の放射性セシウムスクリーニング法」（平成23年11月10日事務連絡）において、所定の測定性能条件を満たせばスクリーニング法の測定装置として使用が認められているNaI-γ線測定装置は、装置管理、測定操作等においては、上に述べたGe測定装置と共通する部分が多いのですが、Ge-γ線測定装置がゲルマニウム半導体中でγ線により生じた電荷を測定するのに対して、NaI-γ線測定装置はヨウ化ナトリウム結晶中でγ線により生じた光を測定するという測定原理の違いから、Ge-γ線測定装置の方が定性、定量能力ともに優れています。今後、食品中放射性物質の規制値が現行暫定規制値の5分の1になるとすれば、NaI-γ線測定装置ではスクリーニング法の測定装置として適さなくなることも想定されます。Ge-γ線測定装置が1台あたりの重量が2トン前後あるため、特殊構造施設が必要であること、その価格が2,000万円以上であるのに対して、NaI-γ線測定装置は設置が容易であること、価格が300～500万円程度であることから、NaI-γ線測定装置は試験検査機関に多数導入され、スクリーニングに活躍しましたが、Ge-γ線測定装置については現存台数のみで食品すべての検査に対応できるほど増えてはおりません。

まとめ

食品中の放射性物質に対する安全確保のための要点を、試験検査を担当する者の立場から説明させて頂きました。食品中の有害物質に対する規格基準は、規格基準自体が科学的根拠に裏付けられて、誰でもが納得できるものであることはもちろん、生産、流通、消費の各段階で適正な管理がなされていなければなりません。リスクマネージメントで重要なポイントである現状把握のための検査体制の拡充が望まれます。信頼性の高い検査結果から、安心を広げたい、復興に役立ちたいと熱望しています。



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CHAPTER 4

Japan Food Allergen Labeling Regulation—History and Evaluation

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Abstract

According to a national survey of food allergy cases, the food-labeling system for specific allergenic ingredients (i.e., egg, milk, wheat, buckwheat, and peanut) in Japan was mandated under law on April 1, 2002. By Japanese law, labeling of allergens is designated as mandatory or recommended based on the number of cases of actual illness and the degree of seriousness. Mandatory labeling is enforced by the ministerial ordinance, and the ministerial notification recommends that foods containing walnut and soybean be labeled with subspecific allergenic ingredients. Additional labeling of shrimp/prawn and crab has also become mandatory since 2008. To monitor the validity of the labeling system, the Japanese government announced the official methods for detection of allergens in a November 2002 ministry notification. These official methods, including two kinds of enzyme-linked immunosorbent assay kits for screening, Western blotting analyses for egg and milk, and polymerase chain reaction analyses for wheat, buckwheat, peanut, shrimp/prawn and crab as confirmation tests, have provided a means to monitor the labeling system. To standardize the official methods, the Japanese government described the validation protocol criteria in the 2006 official guidelines. The guidelines stipulate that any food containing allergen proteins at greater than 10 mg/kg must be labeled under the Law. This review covers the selection of the specific allergenic ingredients by the Japanese government, the implementation of regulatory action levels and the detection methods to support them, and the assessment of the effectiveness of this approach.

I. ASSESSMENT OF IMMEDIATE-TYPE FOOD ALLERGIES IN JAPAN

Food allergies that cause immediate reactions had already been under investigation prior to any discussion of "allergy food labeling" under the food sanitary law for prepackaged processed foods and food additives. Before implementation of the allergy food-labeling system in Japan, a research group supported by the Ministry of Health and Welfare of Japan had collected epidemiological data on immediate-type food allergies during both childhood and adulthood in Japan in 1998 and 1999. This retrospective study asked hospitals with more than 200 beds to report all immediate-type food allergy cases treated by the emergency department. The questionnaire included information on age, sex, cause of the food allergy, symptoms, IgE CAP RAST, and type of treatment. To focus on the

immediate-type, only cases in which symptoms occurred within 60 min after ingestion of the suspected food were included. Of the 2623 hospitals surveyed, 1623 hospitals responded and 1420 cases were analyzed. As shown in Table 4.1, hen's eggs were the most common allergen, followed by cow's milk, wheat, buckwheat, fishes, fruits, and shrimp. The top three major food allergens were most prevalent among the pediatric population, whereas fishes, buckwheat, and shrimp were mainly reported in adults. Based on these data, the Ministry of Health and Welfare selected 24 candidates that caused more than four cases of adverse reaction for the allergy food-labeling system. Following roundtable discussions among specialists and regulatory officers of the Ministry of Health and Welfare, hen's eggs, cow's milk, wheat, buckwheat, and peanuts were selected as items for mandatory labeling by the 2000 ministerial ordinance; the remaining 19 allergens were designated as items for recommended labeling by a ministerial notification.

To further understand the real-time condition of food allergies in Japan, we investigated prospectively the immediate-type food allergy cases in collaboration with more than 2000 doctors between 2001 and 2002 to account for recall bias in the previous study. The contributing doctors included those working in hospitals with more than 200 beds as well as allergy specialists working in clinics. Contributing doctors were asked to respond to a questionnaire every 3 months for 2 years from 2001 to 2002 and report immediate-type food allergy cases by mail. The same questionnaire as that in the previous studies was used, and only immediate-type food allergies as defined in the previous study were included. A total of 3882 cases were reported within the 2 years (Table 4.2). The cases ranged from 0 to 80 years of age, with 50% (1969) of them below 2 years of age. The most common cause of food allergy was hen's eggs (38.3%), followed by cow's milk (15.9%), wheat (8%), shellfish (6.2%), fruits (6%), buckwheat (4.6%), fishes (4.4%), and peanuts (2.8%). Notably, the cause of food allergy differed greatly among age groups. Food-induced anaphylaxis was seen in 10.9% of the reported cases. As shown in Table 4.3, hen's eggs, cow's milk plus its products, wheat, buckwheat, and peanuts were the major causes of food-induced anaphylaxis in Japan. Compared to our previous investigation, fruit allergies against kiwi and banana seemed to be an increasing trend. Thus, the present Ministry of Health, Labor, and Welfare of Japan (MHLW) has been implementing countermeasures against food allergies to improve the quality of life of afflicted patients. This prospective investigation on immediate-type food allergies has been repeated every 3 years as a means to monitor the condition of food allergies in Japan. The results of these investigations have improved the allergy food-labeling system by including banana as a recommended item by a ministerial notification and shrimp and crab as mandatory items for labeling by a ministerial ordinance.

TABLE 4.1 Immediate type of food allergy cases reported from 1998 to 1999

Offending food, *n* (%)	Total	>1 year	1 year	2–3 years	4–6 years	7–19 years	20+ years
Egg	420 (29.6)	197 (47.4)	72 (30.4)	89 (30.8)	35 (25.0)	19 (9.2)	8 (6.1)
Milk product	324 (22.8)	128 (30.8)	66 (27.8)	70 (24.2)	34 (24.3)	21 (10.1)	5 (3.8)
Wheat	147 (10.4)	40 (9.6)	20 (8.4)	35 (12.1)	12 (8.6)	27 (13.0)	13 (9.9)
Buckwheat	82 (5.8)	1 (0.2)	10 (4.2)	16 (5.5)	10 (7.1)	29 (14.0)	16 (12.2)
Fish	73 (5.1)	15 (3.6)	9 (3.8)	10 (3.5)	5 (3.6)	13 (6.3)	21 (16.0)
Fruits	66 (4.6)	6 (1.4)	13 (5.5)	13 (4.5)	8 (5.7)	19 (9.2)	7 (5.3)
Shrimp	51 (3.6)	0 (0.0)	2 (0.8)	4 (1.4)	4 (2.9)	22 (10.6)	19 (14.5)
Meat	44 (3.1)	9 (2.2)	2 (0.8)	4 (1.4)	4 (2.9)	14 (6.8)	11 (8.4)
Peanut	34 (2.4)	3 (0.7)	12 (5.1)	5 (1.7)	6 (4.3)	5 (2.4)	3 (2.3)
Soybean	27 (1.9)	5 (1.2)	8 (3.4)	4 (1.4)	3 (2.1)	4 (1.9)	3 (2.3)
Other	152 (10.7)	12 (2.9)	23 (9.7)	39 (13.5)	19 (13.6)	34 (16.4)	25 (19.1)
Total	1420	416	237	289	140	207	131

TABLE 4.2 Immediate type of food allergy cases reported from 2001 to 2002

Offending food, *n* (%)	Total	>1 year	1 year	2–3 years	4–6 years	7–19 years	+20 years
Egg	1486 (38.3)	789 (62.1)	312 (44.6)	179 (30.1)	106 (23.3)	76 (15.2)	24 (6.6)
Milk product	616 (15.9)	255 (20.1)	111 (15.9)	117 (19.7)	84 (18.5)	41 (8.2)	8 (2.2)
Wheat	311 (8.0)	90 (7.1)	49 (7.0)	46 (7.7)	24 (5.3)	48 (9.6)	54 (14.8)
Fruits	232 (6.0)	40 (3.1)	30 (4.3)	30 (5.1)	40 (8.8)	45 (9.0)	47 (12.8)
Buckwheat	179 (4.6)	4 (0.3)	23 (3.3)	45 (7.6)	27 (5.9)	54 (10.8)	26 (7.1)
Fish	171 (4.4)	21 (1.7)	32 (4.6)	22 (3.7)	18 (4.0)	37 (7.4)	41 (11.2)
Shrimp	161 (4.1)	4 (0.3)	10 (1.4)	20 (3.4)	29 (6.4)	59 (11.8)	39 (10.7)
Peanut	110 (2.8)	4 (0.3)	22 (3.1)	31 (5.2)	28 (6.2)	22 (4.4)	3 (0.8)
Soybean	76 (2.0)	22 (1.7)	16 (2.3)	9 (1.5)	8 (1.8)	9 (1.8)	12 (3.3)
Meat	71 (1.8)	13 (1.0)	6 (0.9)	7 (1.2)	7 (1.5)	19 (3.8)	19 (5.2)
Other	469 (12.1)	28 (2.2)	88 (12.6)	88 (14.8)	83 (18.3)	89 (17.8)	93 (25.4)
Total	3882	1270	699	594	454	499	366

TABLE 4.3 Anaphylaxis cases reported from 2001 to 2002

No.	Offending food	*n* (%)
1	Egg	109 (27.6)
2	Milk product	93 (23.5)
3	wheat	70 (17.7)
4	Buckwheat	28 (7.1)
5	Peanuts	18 (4.6)
6	Shrimp	14 (3.5)
7	Salmon roe	8 (2.0)
	Peach	8 (2.0)
9	Soybean	7 (1.8)
	Kiwi	7 (1.8)
11	Banana	4 (1.0)
	Yam	4 (1.0)
–	Other	25 (6.3)
	Total	395

II. JAPANESE FOOD ALLERGY-LABELING SYSTEM

Food allergies represent an important health problem in industrialized countries. In Japan, the number of people with food allergies is increasing, especially among young children, due to major changes in dietary habits with the introduction of western foods after World War II.

In 1999, the Joint FAO/WHO Codex Alimentary Commission Session agreed to recommend labeling of eight kinds of food which contain ingredients known to be allergens. This movement has led the Japanese government to take new measures to tackle food allergies in Japan.

A. Japanese regulations for labeling of food allergenic ingredients

The special subcommittee of MHLW held a meeting on the labeling of the Food Sanitation Investigation Council and stated that, "From the viewpoint of preventing the occurrence of health hazards, mandatory labeling of foods containing specific allergenic ingredients should be required." Accordingly, the MHLW decided that the Food Sanitation Law should provide for the mandatory labeling of foods containing allergenic ingredients designated in the 2000 ministerial ordinance.

Since the only therapy for a food allergy is avoidance of the responsible food, it is essential for food allergy patients to eliminate food allergens from their diet. Therefore, the Japanese MHLW decided to improve the

allergen-labeling system by amending the Food Sanitation Law in 2001 (Ebisawa *et al.*, 2003). They organized a labeling study group consisting of clinical experts, patients, researchers, retailers, and food industrialists. The group discussed different labeling system methods. The results were announced as a report. In the report outline, labeling was divided into two stages, mandatory and recommended, based on the number of cases of actual illnesses and the degree of seriousness (Table 4.4). Consequently, eggs, milk, wheat, buckwheat and peanuts, and most recently shrimp and crab require mandatory labeling by the ministerial ordinance; hereinafter, we refer to these seven ingredients as "specific allergenic ingredients." In addition, the ministerial notification recommends labeling of any food that contains the following 18 ingredients: abalone, squid, salmon roe, orange, kiwifruit, beef, walnut, salmon, mackerel, soybean, chicken, banana, pork, Matsutake mushroom, peach, yam, apple, and gelatin. Hereinafter, we refer to these ingredients as "subspecific allergenic ingredients." To the best of our knowledge, Japan is the first country to set up mandatory food allergy labeling and to regulate it under national law in 2002. The additional labeling requirement for shrimp/prawn and crab was introduced by the amendment of the food Sanitation Law under the MHLW in June 2008 due to the almost unlimited use of crustaceans in processed foods in Japan and the frequency of adverse food reactions in allergic patients.

Among shrimp allergy cases, 64.7% of patients showed positive reaction to crabs. The clinical evidence suggests that many shrimp allergy patients react to crabs. On the contrary, as the remaining 35.3% of patients showed no reaction to crabs, some patients with shrimp allergy can eat crabs. Thus, it would be important to label "shrimp" and "crab" separately, rather than as "crustacean" to give consumers more information. Accordingly, the MHLW has revised the mandatory labeling for shrimp

TABLE 4.4 Allergenic ingredients designated by the MHLW of Japan*

Specific allergenic ingredients
Mandatory by ministerial ordinance (seven ingredients)
Egg, milk, wheat, buckwheat, peanut, shrimp/prawn, and crab
Subspecific allergenic ingredients
Recommended by ministerial notification (18 ingredients)
Abalone, squid, salmon roe, orange, kiwifruit, beef, walnut, salmon, mackerel, soybean, chicken, banana, pork, Matsutake mushroom, peach, yam, apple, and gelatin

*Based on the Notification of March 15, 2001 and the newest Notification of June 3, 2008 from the Department of Food Safety, Ministry of Health, Labor, and Welfare (MHLW) of Japan.

and crab to be labeled separately. Since the management of the food-labeling policy was transferred from the MHLW to the Consumer Affairs Agency (CAA) in 2010, CAA announces the Japanese food-labeling system through ministry notifications.

The content scope of allergens for labeling was established based on the Japan Standard Commodity Classification. Japan is the first country to set up mandatory food allergy labeling and to regulate it under national law.

The characteristics of the Japanese labeling system are as follows.

1. Small quantity labeling

The specific allergenic ingredients must be labeled even in cases of carry-over conditions or when used as processing aids. Labeling of the 18 subspecific allergenic ingredients in Table 4.1 is recommended as much as possible.

2. "May contain" labeling

"May contain (name of allergenic ingredients)" type labeling is prohibited.

3. Combination of specified ingredients

With a few exceptions, the use of major item classifications (declaration of meats, cereals, etc.) is prohibited.

4. Declaration of high-grade food ingredients

In cases with high-grade food ingredients such as abalone, salmon roe, and mushroom mixed in very small quantities, a declaration such as "contains xxx extract" is required so as not to mislead consumers.

5. Method of declaring additives

For food additives, labeling shall, in principle, declare the "name of the substance (derived from)."

6. Declaration of flavorings

Aromatic ingredients have not yet been subjected to labeling, but should be labeled as much as possible.

7. Alcoholic beverages and related products

Alcoholic beverages are not currently subject to regulated labeling.