Provisional Translation
from the Japanese Original

Specifications and Standards for Food, Food Additives, Etc.

Ministry of Health and Welfare Notification No. 370, 1959

The Minister of Health and Welfare has established the specifications and standards for food, food additives given below, based on Article 10 and Article 11 Paragraph 1 of the Food Sanitation Law (Law No. 233, 1947).

Part I Food

Section A General Compositional Standards for Food

1. Foods shall not contain any antibiotics or chemically synthesized antibacterial substances (substances obtained by instigating chemical reactions to elements and/or compounds through chemical methods, except for decomposition; this applies hereinafter in this paragraph), except for the following cases:

(1) When the substance concerned is identical to the food additive determined by the Minister of Health, Labour and Welfare as having no potential to cause damage to human health under Article 10 of the Food Sanitation Law (Law No. 233, 1947, hereinafter “the Law.”)

(2) When compositional standards are set forth in 5, 6, 7, 8 or 9 below for the substance concerned.

(3) When the food product concerned has been manufactured or processed using a food ingredient that meets the compositional standards given in 5, 6, 7, 8 or 9 below (except for foods containing antibiotics or chemically synthesized antibacterial substances for which compositional standards are not set forth in 5, 6, 7, 8 or 9 below.)

2.~4. (Omitted)

5. Foods shall not contain substances (including substances produced by chemical transformation: this applies hereinafter in this paragraph) used as ingredients of agricultural chemicals and other chemical substances listed in the table in (1) below. The agricultural chemicals and other chemical substances stated above, here and also later in this paragraph, refer to substances used for purposes designated by the Agriculture, Forestry and Fisheries Ministerial Ordinance according to the provision

of Paragraph 3 of Article 2 of the Law Concerning Safety Assurance and Quality Improvement of Agricultural Chemicals and Feeds (Law No. 35, 1953), which is stipulated under Paragraph 1 of Article 1-2 of the Agricultural Chemicals Regulation Law (Law No. 82, 1948), with such aims as adding to, mixing with, or infiltrating into feeds (the feeds stipulated under Paragraph 2 of Article 2 of Law No. 35), or medical products to be used for animals, which are stipulated in Paragraph 1 of Article 2 of the Pharmaceutical Affairs Law (Law No. 145, 1960). In association with this regulation, a sample of the foods listed in the "foods" column in the table in (2) below shall be tested using the part listed in the "samples" column in the table by the testing methods described in (3) to (16) below. No ingredients of agricultural chemicals or other chemical substances shall be detected in these tests.

(1) Substances used as ingredients of agricultural chemicals and other chemical substances that are stipulated to be "Not detected" in foods

1. 2, 4, 5-T
2. Azocyclotin and cyhexatin
3. Amitrol
4. Captafol
5. Carbadox
6. Coumaphos
7. Chloramphenicol
8. Chlorpromazine
9. Diethylstilbestrol
10. Dimetridazole
11. Daminozide
12. Nitrofurans
13. Propham
14. Malachite green
15. Metronidazole
16. Ronidazole

(2) Samples

Food	Sample material
Barley and buckwheat	Threshed seeds
Wheat and rye	Husked seeds
Rice (brown rice)	Husked seeds
Corn (maize)	Seeds with the husks, the silk and the cores removed
Other cereal grains	Threshed seeds
Peas, beans (dry)(including butter beans, cowbeans (red beans), lentil, lima beans, pegia, sultani, sultapya and white beans)*, broad beans and soybeans (dry))	Seeds without the pods
Peanuts	With the shells removed
Other legumes/pulses	Seeds without the pods
Apricot, mume plum, cherry, Japanese plum (including prune) and nectarine	With the peduncle and the seeds removed
Peach	With the skins and the seeds removed
Orange (including navel orange), grapefruit, citrus *natsudaidai* (whole), lime and lemon	Whole fruit
Citrus *natsudaidai* (pulp) and *unshu* orange (pulp)	With the peels removed
Citrus *natsudaidai*, peels	With the calyxes removed
Other citrus fruits	Whole fruit
Pear, Japanese pear, quince and apple	With the blossom scars, the cores and the peduncles removed
Loquat	With the peduncles, the skins and the seeds removed
Avocado and mango	With the seeds removed
Kiwifruit	With the skins removed
Guava	With the calyxes removed
Date	With the calyxes and the seeds removed

Pineapple	With the tops removed
Passion fruit and papaya	Whole fruit
Banana	With the pedicels removed
Strawberry, cranberry, huckleberry, blackberry and blueberry	With the calyxes removed
Raspberry	Whole fruit
Other berries	With the calyxes removed
Japanese persimmon	With the calyxes and the seeds removed
Watermelon, *makuwauri* melon and melons	With the rinds removed
Grape	With the peduncles removed
Other fruits	Edible portions
Turnip (roots) and Japanese radish (roots, including radish)	With the dirt lightly rinsed off with water
Turnip (leaves), watercress, kale, Japanese radish (leaves, including radish), and brussels sprouts	With the decayed leaves removed
Cauliflower and broccoli	With the leaves removed
Cabbage and Chinese cabbage	A sample consisting of one portion from each of four heads, with each head equally cut into four portions, without the decayed outer leaves and the cores
Kyona and *komatsuna* (Japanese mustard spinach)	With the roots and the decayed leaves removed
Horseradish	Roots with the dirt lightly rinsed off with water
Qing-geng-cai and other cruciferous vegetables	Edible portions
Sweet potato, konjac, taro, potato, yam and other potatoes	With the dirt lightly rinsed off with water
Pumpkin (including squash), cucumber (including gherkin) and oriental pickling melon (vegetable)	With the vines removed
Other cucurbitaceous vegetables	Edible portions
Artichoke, endive and chicory	With the decayed leaves removed

Burdock and salsify	A sample thinly sliced then ground with a meat grinder, the leaves having been removed and the dirt having been lightly rinsed off with water
Shungiku	With the roots and the decayed leaves removed
Lettuce (including cos lettuce and leaf lettuce)	With the decayed outer leaves and the cores removed
Other composite vegetables	Edible portions
Shiitake mushroom, button mushroom and other mushrooms	Edible portions
Celery, parsley and *mitsuba*	With the roots and the decayed leaves removed
Carrot and parsnip	With the dirt lightly rinsed off with water
Other umbelliferous vegetables	Edible portions
Tomato, egg plant and pimiento (sweet pepper)	With the calyxes removed
Other solanceous vegetables	Edible portions
Asparagus	Stems
Onion, garlic, welsh (including leek) and multiplying onion	With the outer skins and the root hair removed
Nira and other liliaceous vegetables	Edible portions
Green soybeans, kidney beans (immature, with pods) and peas (immature, with pods)	With the pedicels removed
Okra	With the calyxes removed
Sugarcane	With the husks removed
Ginger	With the leaves removed, and with the dirt lightly rinsed off with water
Sugar beet	With the dirt lightly rinsed off with water
Spinach	With red roots left on, and with the root hair and the decayed leaves removed
Bamboo shoots and other vegetables	Edible portions

Sesame seeds, rapeseeds, sunflower seeds, safflower seeds, cotton seeds and other oil seeds	Seeds only
Almond, ginkgo nut, chestnut, walnut, pecan and other nuts	With the husks removed
Cacao beans and coffee beans	Beans without the pods
Tea	Tea leaves
Hop	Dried flowers
Other spices and other herbs	Edible portions

(3) 2, 4, 5-T analytical method

1. Apparatus

A gas chromatograph with an electron capture detector (GC-ECD) and a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS).

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Ether: Use a rotary vacuum evaporator on 300 mL of ether to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is found to include any substance that may interfere with the analysis of

substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Synthetic magnesium silicate (Florisil) for column chromatography: Heat florisil (150-250 μm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Diatomaceous earth: Diatomaceous earth for chemical analysis..

Ethyl acetate: Use a rotary vacuum evaporator on 300 mL of ethyl acetate to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

Reagent for butyl esterification: Dissolve 10 g of boron trifluoride ether complex in 25 ml of *n*-butanol.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Methanol: Use a rotary vacuum evaporator on 300 mL of methanol to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

3. Reference standard

2,4,5-T: This product contains not less than 98% of 2,4,5-T, and its melting point is 156°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses and seeds

Weigh 10.0 g of the test sample, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for two hours.

Then, add 100 ml of acetone and 5 ml of 4 mol/l hydrochloric acid, and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper. Add 50 ml of acetone and homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of ethyl acetate and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave the funnel to stand, and then transfer the ethyl acetate layer to a 300-ml conical flask. Add 50 ml of ethyl acetate to the aqueous layer and repeat the above procedure, and then add the ethyl acetate layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of ethyl acetate and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and concentrate the mixture to approximately 1 ml at 40°C or lower, and then evaporate the solvent to near dryness in a nitrogen stream at room temperature.

Add 30 ml of *n*-hexane to the dried residue and transfer the mixture to a 100-ml separating funnel, and then add 30 ml of *n*-hexane-saturated acetonitrile. Shake the mixture vigorously for five minutes using a shaker and leave the funnel to stand, and then transfer the acetonitrile layer to a 200-ml separating funnel. Add 30 ml of *n*-hexane-saturated acetonitrile to the *n*-hexane layer and repeat the above procedure twice, and then combine the acetonitrile

layer to the separating funnel above. Add 50 ml of *n*-hexane-saturated acetonitrile into the separating funnel and shake lightly, and then leave the funnel to stand. Transfer the acetonitrile layer into a rotary vacuum evaporator and concentrate the mixture to approximately 1 ml at 40°C or lower, and then evaporate the solvent to near dryness in a nitrogen stream at room temperature.

ii. Fruit, vegetables, matcha and hops

In case of fruit and vegetables, weigh accurately about 1 kg of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and mesure out a sample equivalent to 20.0 g.

In case of matcha, weigh 5.00 g of the test sample and add 20 ml of water, and then leave it to stand for two hours.

In case of hops, weigh 5.00 g of the test sample, previously ground, and add 20 ml of water, and then leave it to stand for two hours.

Then, add 100 ml of acetone and 5 ml of 4 mol/l hydrochloric acid, and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of ethyl acetate and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave the funnel to stand, and then transfer the ethyl acetate layer to a 300-ml conical flask. Add 50 ml of ethyl acetate to the aqueous layer and repeat the above procedure twice, and then combine the ethyl acetate layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of ethyl acetate and wash twice the residue on the

surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and concentrate the mixture to approximately 1 ml at 40°C or lower, and then evaporate the solvent to near dryness in a nitrogen stream at room temperature.

iii. Teas other than matcha

Weigh 9.00 g of the test sample and soak it in 540 ml of water at 100°C, and then leave it to stand at room temperature for five minutes. Transfer 360 ml of the cooled filtrate into a 500-ml conical flask and add 18 g of sodium chloride and 4 mol/l hydrochloric acid to adjust the pH to 1 or lower. Transfer the solution to a 1,000-ml separating funnel already containing 100 ml of ethyl acetate. Shake the mixture vigorously for five minutes using a shaker and leave the funnel to stand, and then transfer the ethyl acetate layer to a 300-ml conical flask. Add 100 ml of ethyl acetate to the aqueous layer and repeat the above procedure twice, and combine the ethyl acetate layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of ethyl acetate and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and concentrate the mixture to approximately 1 ml at 40°C or lower, and then evaporate the solvent to near dryness in a nitrogen stream at room temperature.

iv. Foods other than those listed in i to iii above

Obtain the extracts according to the methods described in i or ii.

b. Hydrolysis

Dissolve the residue obtained by the extraction described in 4-a in 20 ml of methanol and transfer the mixture to a 100-ml eggplant-shaped flask, and then add 10 ml of 1.5 mol/l sodium hydroxide solution. Attach a reflux condenser to the flask and heat the mixture for 30 minutes in a water bath at 80°C, and then allow it to cool. Transfer the solution into a rotary vacuum evaporator and remove most of the methanol at 40°C or lower. Filter the residue by suction into a 300-ml separating funnel (I) through a glass filter (pore size G3). Wash the residue on the glass filter with a small amount of acetone and water and add the washings to the separating funnel above, and then add 50

ml of ether and 100 ml of 10% sodium chloride solution. Shake the mixture vigorously for five minutes using a shaker and leave it to stand. Transfer the aqueous layer to a 300-ml separating funnel (II) and add 4 mol/l hydrochloric acid to adjust the pH to 1 or lower. Add 50 ml of ethyl acetate to the adjusted solution and shake the mixture vigorously for five minutes using a shaker, and then leave it to stand. Transfer the ethyl acetate layer to a 300-ml conical flask. Add 50 ml of ethyl acetate to the aqueous layer and repeat the above procedure, and then combine the ethyl acetate layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of ethyl acetate and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and concentrate the mixture to approximately 1 ml at 40°C or lower.

c. Butyl esterification

Transfer the solution obtained by the hydrolysis described in 4-b to a 20-ml eggplant-shaped flask and then evaporate the solvent to near dryness in a nitrogen stream at room temperature. After the desiccation, add 1 ml of a butyl esterificated agent to the residue. Attach a reflux condenser to the eggplant-shaped flask described above and heat the mixture for 30 minutes in a water bath at 90°C, and allow it to cool. Transfer the mixture to a 200-ml separating funnel already containing 50 ml of 10% sodium chloride solution and 50 ml of *n*-hexane and shake the mixture vigorously for five minutes using a shaker, and then leave it to stand. Transfer the *n*-hexane layer to a 200-ml conical flask and add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 10 ml of *n*-hexane and wash the residue on the surface of the filter paper with the washings. Add the washings to the rotary vacuum evaporator and concentrate the mixture to approximately 2 ml at 40°C or lower.

d. Clean-up

Add 5 g of florisil for column chromatography suspended in *n*-hexane into a chromatograph tube (15mm in inner diameter, 300 mm in length) and add over approximately 5 g of sodium sulfate (anhydrous) into the column. Spill out the *n*-hexane until only a small amount remains on the packing of the column and pour the solution obtained by the butyl esterification described in 4-c into the column. Pour 50 ml of ether/*n*-hexane (1:19) into the column and discard the effluent. Pour 150 ml of ether/*n*-hexane (3:17) into the column and collect the eluate into a rotary vacuum evaporator. Concentrate the mixture to approximately 1 ml at 40°C or lower and evaporate the solvent to near dryness in a nitrogen stream at room temperature. Dissolve the residue in *n*-hexane to make exactly 10 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard under the procedure described in 4-c.

Testing conditions

Column: A silicate glass capillary column (0.25 mm in inner diameter, 30 m in length) coated with 5% phenyl methyl silicone for gas chromatography to a thickness of 0.25 μm.

Column temperature: Hold the column temperature at 50°C for one minute, followed by an increase of 25°C every minute until reaching 125°C, after which increase the temperature by 10°C every minute until reaching 300°C, and hold for five minutes.

Inlet temperature: 260°C

Detector: Operate at 300°C.

Gas flow rate: Use nitrogen or helium as the carrier gas. Adjust the flow rate so that *n*-butyl (2,4,5-trichlorophenoxy) acetate flows out in approximately 15 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the same

conditions as those described in 5-a. Test results obtained for the reference standard must be the same as those obtained under the procedure described in 4-c. Determine the quantity using either the peak height or peak area method, if required.

(4) Analytical method for azocyclotin and cyhexatin

1. Apparatus

A gas chromatograph with a flame photometric detector (GC-FPD, interference filter for tin determination, wavelength: 610nm) and a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS).

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

3 mol/l ethylmagnesium bromide-ethereal solution: 3 mol/l ethyl magnesium bromide-ethereal solution.

Ether: Use a rotary vacuum evaporator on 300 mL of ether to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is found to include any substance that may interfere with the analysis of substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Synthetic magnesium silicate (Florisil) for column chromatography:

Heat florisil (150-250 μm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

Cyhexatin standard solution: Add acetic acid/ethyl acetate (1:99) to 10.0 mg of cyhexatin to make a 100-ml solution. Take out 10 ml of the solution and add *n*-hexane to make 100 ml of mix.

Sodium dodecyl sulfate: Sodium dodecyl sulfate with a purity of 85% or higher.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

3. Reference standard

Cyhexatin: This product contains not less than 99% of cyhexatin, and its melting point is 195-198°C.

4. Procedure

a. Extraction

i. Legumes/pulses and seeds

Weigh 10.0 g of the test sample, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for two hours.

Then, add 100 ml of acetone/acetic acid (99:1) and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone/acetic acid (99:1), and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum

evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Trasfer the concentrated solution to a 500-ml separating funnel already containing 200 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then add the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower.

Add 20 ml of *n*-hexane to the residue and transfer the mixture to a 100-ml separating funnel, and then add 40 ml of n-hexane-saturated acetonitrile. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the acetonitrile layer into the rotary vacuum evaporator. Add 40 ml of *n*-hexane-saturated acetonitrile to the *n*-hexane layer and repeat the above procedure twice, and then combine the acetonitrile layer into the rotary vacuum evaporator above. Remove the acetonitrile at 40°C or lower.

Dissolve the residue in *n*-hexane to make exactly 5 ml of solution.

ii. Cereal grains, fruit and vegetables

In case of cereal grains, weigh 10.0 g of the test sample, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for two hours.

In case of fruit and vegetables, weigh accurately 1 kg of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and measure out a sample equivalent to 20.0 g.

Then, add 100 ml of acetone/acetic acid (99:1) and homogenize the mixture for three minutes. Filter the homogenized sample by suction

into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone/acetic acid (99:1), and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 500-ml separating funnel already containing 200 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure twice, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane,and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower, and then dissolve the residue in *n*-hexane to make exactly 10 ml of solution.

iii. Matcha and hops

In case of matcha, weigh 5.00 g of the test sample and add 20 ml of water, and then leave it to stand for two hours.

In case of hops, weigh 5.00 g of the test sample, previously ground, and add 20 ml of water, and then leave it to stand for two hours.

Then, add 100 ml of acetone/acetic acid (99:1) and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone/acetic acid (99:1), and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 500-ml separating funnel already containing 200 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure twice, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and concentrate the mixture to approximately 5 ml at 40°C or lower, and then add *n*-hexane to make exactly 10 ml of solution.

iv. Teas other than matcha

Weigh 9.00 g of the test sample and soak it in 540 ml of water at 100°C, and then leave it to stand at room temperature for five minutes. Transfrer 360 ml of the cooled filtrate into a 500-ml separating funnel and add 30 g of sodium chloride, 2 ml of 2% sodium dodecyl sulfate solution and 100 ml of *n*-hexane. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 100 ml of *n*-hexane to the aqueous layer and repeat the above procedure twice, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove *n*-hexane at 40°C or lower. Dissolve the residue in *n*-hexane to make exactly 6 ml of solution.

v. Foods other than those listed in i to iv above

Obtain the extracts according to the methods described in i, ii, or

iii.

b. Ethylation

Transfer 1 ml (2 ml for cereal grains, teas and hops) of the solution obtained by the extraction described in 4-a to a 50-ml test tube with a glass stopper and add 1 ml (2 ml for cereal grains, teas and hops) of a 3 mol/l ethylmagnesium bromide-ethereal solution, and then leave it to stand for 20 minutes at room temperature.

Then, add 10 ml of 0.5 ml/l sulfuric acid gradually and add 10 ml of water to be mixed. Add 10 ml of *n*-hexane to the mixture and shake the mixture vigorously for one minute, and then leave it to stand. Transfer the *n*-hexane layer to a 50-ml conical flask. Add 5 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 5 ml of *n*-hexane and wash the residue on the surface of the filter paper with the washings. Add the washings to the rotary vacuum evaporator and concentrate the mixture to 2 ml at 40°C or lower.

c. Clean-up

Add 5 g of florisil for column chromatography suspended in *n*-hexane into a chromatograph tube (15 mm in inner diameter, 300 mm in length) and add over approximately 5 g of sodium sulfate (anhydrous) into the column. Spill out the *n*-hexane until only a small amount remains on the packing of the column and pour the solution obtained by the ethylation described in 4-b into the column. Then, wash the eggplant-shaped flask of the rotary vacuum evaporator with 15 ml of *n*-hexane and transfer rhe washings to the column, and then collect the eluate into the rotary vacuum evaporator. Then, pour 50 ml of ether/*n*-hexane (1:99) into the column and collect the eluate in the rotary vacuum evaporator, and then remove the ether and *n*-hexane at 40°C or lower. Dissolve the residue in *n*-hexane to make exactly 2 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the cyhexatin

reference standard under the procedure described in 4-b. Azocyclotin is modified by ethylation into the same substance as cyhexatin.

Testing conditions

Column: A silicate glass capillary column (0.32 mm-0.53 mm in inner diameter, 30 m in length) coated with 5% phenyl methyl silicone for gas chromatography to a thickness of 1.5 μm.

Column temperature: Hold the column temperature at 120°C for two minutes, followed by an increase of 10°C every minute until reaching 200°C, after which increase the temperature by 20°C every minute until reaching 300°C, and hold for five minutes.

Inlet temperature: 280°C

Detector: Operate at 300°C.

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate so that cyhexatin flows out in approximately 13-15 minutes. Adjust the flow rates of air and hydrogen to optimal conditions.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the same conditions described in 5-a. Test results obtained for the reference standard must be same as those obtained under the procedure described in 4-b. Determine the quantity using either the peak height or peak area method, if required.

(5) Amitrole analytical method

1. Apparatus

A high-performance liquid chromatograph with a fluorescence detector (HPLC-FL) and a liquid chromatograph/mass spectrometer (LC/MS or LC/MS/MS).

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD.

Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Ethanol: Use a rotary vacuum evaporator on 300 mL of ethanol to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 µL of the solution into a gas chromatograph equipped with an electron capture detector. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

Acetic acid buffer solution: Add 0.05 mol/l sodium acetate solution to 800 ml of 0.05 mol/l acetic acid solution to make a 1,000-ml solution.

Weakly acidic cation exchange resin: Wash the weakly acidic cation exchange resin produced for column choromatography with 1 mol/l hydrochloric acid, secondly with a 2.8% aqueous ammonia, and thirdly with 1 mol/l hydrochloric acid again. Thereafter, wash the washings with water until they become neutral.

Fluorescamin: A reagent with a purity of 98% or higher.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Phosphate buffer solution: Add 10% phosphoric acid solution to a 0.05 ml/l monosodium phosphate solution to adjust the pH to 3.0.

3. Reference standard

Amitrole: This product contains not less than 98% of amitrole, and its melting point is 157-159°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses, seeds, fruit, vegetables, matcha and hops

In case of cereal grains, legumes/pulses and seeds, weigh 30.0 g of the test sample, previously ground so as to pass through a standard mesh sieve (420 µm).

In case of fruit and vegetables, weigh accurately about 1 kg.of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and measure out a sample equivalent to 30.0 g.

In case of matcha, weigh 30.0 g of the test sample.

In case of hops, weigh 30.0 g of the test sample, previously ground. Then, add 80 ml of ethanol and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth, and then transfer the filtrate to a 200-ml graduated cylinder. Collect the residue on the surface of the filter paper and add 40 ml of 60% ethanol, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate to the graduated cylinder above, and then measure the amount of filtrate.

Transfer 10 ml of the above filtrate to a 200-ml round bottom flask and add 1 ml of hydrogen peroxide solution. Attach a reflux condenser to the flask and heat the mixture for 30 minutes in a water bath at 75°C, and then allow it to cool.

ii. Teas except matcha

Weigh 10.0 g of test sample and soak it in 600 ml of water at 100°C, and then leave it to stand at room temperature for five minutes. Transfer 12 ml of the cooled filtrate into a 200-ml round bottom flask and add 1 ml of hydrogen peroxide solution. Attach a reflux condenser to the flask and heat the mixture for 30 minutes in a water bath at 75°C, and then allow it to cool.

iii. Foods except those listed in i and ii above

Obtain the extracts according to the methods described in i.

b. Clean-up

Add 1 ml of strongly acidic cation exchange resin (0.063-0.156 μm in particle size) suspended in water into a chromatograph tube (10 mm in inner diameter, 300 mm in length) and discard the water until only a small amount remains on the packing of the column. Pour 5 ml of water into the column and discard the effluent. Pour the solution into the column obtained by the extraction described in 4-a. Then, wash the above round bottom flask with 10 ml of water and transfer the washings into the column, and then discard the effluent. Pour 12 ml of 2.8% aqueous ammonia into the column and collect the eluate in a rotary vacuum evaporator. Add 30 ml of *n*-propanol and remove the water and *n*-propanol at 45°C or lower. Dissolve the residue in 5 ml of water.

Add 5 ml of weakly acidic cation exchange resin (0.33-0.50 µm in particle size) suspended in water into a chromatograph tube (10 mm in inner diameter, 300 mm in length) and discard the water until only a small amount remains on the packing of the column. Pour 10 ml of water into the column and discard the effluent. Pour the above solution into the column and discard the effluent. Pour 50 ml of water into the column and discard the effluent. Pour 35 ml of 2.8% aqueous ammonia into the column and collect the eluate in a rotary vacuum evaporator. Add 100 ml of *n*-propanol and remove the aqueous ammonia and *n*-propanol at 45°C or lower.

c. Derivatization

Add 2 ml of acetic acid buffer solution to the residue to dissolve it well. Add 100 µl of 0.25% fluorescamin-acetone solution to 1 ml of the above solution and shake the mixture well, and then leave it to stand for one hour. Add 0.5 ml of 0.05 mol/l sodium borate solution to use as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard under the procedure described in 4-c.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (5 µm in particle size).

Column: A stainless tube (4.6 mm in inner diameter, 150 mm in length).

Column temperature: 40°C

Detector: Operate with an excitation wavelength of 380 nm and a fluorescent wavelength of 484 nm.

Mobile phase: Use acetonitrile/phosphate buffer solution (3:7). Adjust the flow rate so that amitrole flows out in approximately 15 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform liquid chromatography/mass spectrometry under the same conditions described in 5-a. Test results obtained for the reference standard must be the same as those obtained under the procedure described in 4-c. Determine the quantity using either the peak height or peak area method, if required.

(6) Captafol analytical method

1. Apparatus

A gas chromatograph with an electron capture detector (GC-ECD) and a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS).

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is found to include any substance that may interfere with the analysis of substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Synthetic magnesium silicate (Florisil) for column chromatography: Heat florisil (150-250 μm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

Ethyl acetate: Use a rotary vacuum evaporator on 300 mL of ethyl acetate to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting

chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 µL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

3. Reference standard

Captafol: This product contains not less than 98% of captafol, and its melting point is 159-161°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses and seeds

Weigh 10.0 g of the test sample, previously ground so as to pass through a standard mesh sieve (420 µm). Add 20 ml of 3% phosphoric acid solution to the obtained sample and leave it to stand for two hours.

Then, add 100 ml acetone and homogenize the mixture for three minutes Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane,and add the washings to the separating

funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and add the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower.

Add 30 ml of *n*-hexane to the residue and transfer the mixture to a 100-ml separating funnel, and then add 30 ml of *n*-hexane-saturated acetonitril. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the acetonitrile layer into the rotary vacuum evaporator. Add 30 ml of *n*-hexane-saturated acetonitrile to the *n*-hexane layer and repeat the above procedure twice, and then combine the acetonitrile layer into the rotary vacuum evaporator above. Remove the acetonitrile at 40°C or lower and dissolve the residue in 5 ml of *n*-hexane.

ii. Fruit, vegetables, matcha and hops

In case of fruit and vegetables, weigh accurately 1 kg of the test sample and add 500 ml of 10% phosphoric acid solution. Homogenize the mixture and mesure out a sample equivalent to 20.0 g.

In case of matcha, weigh 5.00 g of the tet sample and add 20 ml of 3% phosphoric acid solution, and then leave it to stand for two hours.

In case of hops, weigh 5.00 g of the test sample, previously ground, and add 20 ml of 3% phosphoric acid solution, and then leave it to stand for two hours.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Rpeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate

the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure twice, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower. Dissolve the residue in 5 ml of *n*-hexane.

iii. Teas except matcha

Weigh 9.00 g of the test sample and soak it in 540 ml of water at 100°C, and then leave it to stand at room temperature for five minutes. Transfer 360 ml of the cooled filtrate into a 500-ml conical flask.

Add 30 ml of phosphoric acid, 100 ml of acetone and 2 ml of saturated lead acetate solution to the above conical flask, and then leave the mixture to stand for one hour at room temperature. Filter the mixture by suction into a 1,000-ml separating funnel through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Wash the above conical flask with 50 ml of acetone and wash the residue on the surface of the filter paper with the washings. Add the washings into the above separating funnel and also add 30 g of sodium chloride and 100 ml of *n*-hexane. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 100 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then add the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the

content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower. Dissolve the residue in 5 ml of *n*-hexane.

iv. Foods except those listed in i to iii above

Obtain the extracts according to the methods described in i or ii.

b. Clean-up

Add 5 g of florisil for column chromatography suspended in *n*-hexane into a chromatograph tube (15 mm in inner diameter, 300 mm in length) and add over approximately 5 g of sodium sulfate (anhydrous) into the column. Spill out the *n*-hexane until only a small amount remains on the packing of the column and pour the solution obtained by the extraction described in 4-a into the column. Then, pour 100 ml of *n*-hexane into the column and discard the effluent. Pour 150 ml of ethyl acetate/*n*-hexane (1:9) into the column and collect the eluate into a rotary vacuum evaporator, and then romove the ethyl acetate and *n*-hexane at 40°C or lower. Dissolve the residue in *n*-hexane to make exactly 5 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions 1

Column: A silicate glass capillary column (0.25 mm inner diameter, 10-30 m in length) coated with methyl silicone for gas chromatography to a thickness of 1.5 μm.

Column temperature: Hold the column temperature at 50°C for one minute, followed by an increase of 25°C every minute until reaching 175°C, after which increase the temperature by 10°C every minute until reaching 300°C, and hold for five minutes.

Inlet temperature: 230°C

Detector: Operate at 300°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate to the optimal condition.

Testing conditions 2

Column: A silicate glass capillary column (0.25 mm in inner diameter, 10-30 m in length) coated with 5% phenyl methyl silicone for gas chromatography to a thickness of 0.25 μm.

Column temperature: Hold the column temperature at 50°C for one minute, followed by an increase of 25°C every minute until reaching 125°C, after which increase the temperature by 10°C every minute until reaching 300°C, and hold for three minutes.

Inlet temperature: 230°C

Detector: Operate at 300°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate to the optimal condition.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the same conditions described in 5-a. Test results obtained must be the same as those obtained for the reference standard. Determine the quantity using either the peak height or peak area method, if required.

(7) Carbadox analytical method

Analyze quinoxaline-2-carboxylic acid.

1. Apparatus

A high-performance liquid chromatograph with an ultraviolet spectrophotometric detector (HPLC-UV) and a liquid chromatograph/ mass spectrometer (LC/MS or LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as “special grade” in this section must meet the requirements for “special grade” specified in the Japan Industrial Standards for the reagents.

Octadecylsilane-bonded silica gel cartridge column (500 mg): A polyethylene column of 8-9 mm in inner diameter packed with 500 mg of octadecylsilane-bonded silica gel, or a column equivalent to the specified one in separation capability.

Strongly basic anion exchanger cartridge column (360 mg): A

polyethylene column of 8-9 mm in inner diameter packed with 360 mg of trimethylamino acrylamide copolymer silane-bonded silica gel, or a column equivalent to the specified one in separation capability.

Water: Water produced for liquid chromatography.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Phosphate buffer solution (pH 2.5): Dissolve monopotassium dihydrogen monophosphate (1.36 g) in water to make a 800-ml solution, and add phosphoric acid to adjust the pH to 2.5. Add water to the adjusted solution to make a 1,000-ml solution.

3. Reference standard

Quinoxaline-2-carboxylic acid: This product contains not less than 99% of quinoxaline-2-carboxylic acid, and its melting point is 208°C.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground.. For muscle, remove the fat layer as much as possible before grinding.

Then, add 100 of methanol/0.3% metaphosphoric acid solution (3:7) and homogenize the mixture. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 2-mm thick layer of diatomaceous earth. Wash the residue on the surface of the filter paper with 10 ml of methanol/0.3% metaphosphoric acid solution (3:7) and filter the washings,into the rotary vacuum evaporator. Concentrate the mixture to 30 ml at 45°C or lower and add 0.1 ml of phosphoric acid.

b. Clean-up

i. Octadecylsilane-bonded silica gel column chromatography

Add 5 ml of methanol to an octadecylsilane-bonded silica gel cartridge column (500 mg) followed by 10 ml of water and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 20 ml of phosphate buffer solution (pH 2.5) and discard the effluent. Pour 10 ml of methanol into the column and collect the eluate in a rotary vacuum evaporator, and then remove the methanol at 40°C or lower. Dissolve the residue in 5 ml of water.

ii. Strongly basic anion exchanger column chromatography

Pour 5 ml of water into a strongly basic anion exchanger cartridge column (360 mg) and discard the effluent. Pour the solution obtained in 4-b-i into the column followed by 2 ml of water, 10 ml of ethanol and 5 ml of water, and discard the effluent. Pour 5 ml of 0.1 mol/l hydrochloric acid into the column and collect the eluate in a 50-ml test tube. Add 3 ml of 3 mol/l hydrochloric acid and 15 ml of ethyl acetate and shake the mixture vigorously for five minutes using a shaker, and then leave it to stand. Transfer the ethyl acetate layer into a 100-ml conical flask. Add 15 ml of ethyl acetate to the aqueous layer in the test tube and repeat the above procedure, and then add the ethyl acetate layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator and remove the ethyl acetate at 40°C or lower. Dissolve the residue in 1.0 ml of acetonitrile/phosphate buffer solution (pH 2.5) (1:4), which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (5 µm in particle size).

Column: A stainless tube (4.0-6.0 mm in inner diameter, 150 mm in length).

Column temperature: 40°C

Detector: Operate with an absorption wavelength of 245 nm.

Mobile phase: Use acetonitrile/phosphate buffer solution (pH 2.5) (1:4). Adjust the flow rate so that quinoxaline-2-carboxylic acid flows out in approximately 10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform liquid chromatography/mass spectrometry under the same conditions described in 5-a, but the mobile phase should be acetonitril/water (1:4). Test results obtained must be the same as those obtained for the reference standard. Determine the quantity using either the peak height or peak area method, if required.

(8) Coumaphos analytical method

1. Apparatus

A gas chromatograph with an alkali flame ionization detector (GC-FID), a flame photometric detector (GC-FPD, interference filter for phosphorus determination, wavelength: 526 nm), or a highly-sensitive nitrogen phosphorus detector (GC-NPD), and a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS).

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 µL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 µL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is found to include any substance that may interfere with the analysis of substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Silica gel for column chromatography (63-200 µm in particle size): Heat silica gel made for column chromatography (63-200 µm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

Ethyl acetate: Use a rotary vacuum evaporator on 300 mL of ethyl acetate to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

3. Reference standard

Coumaphos: This product contains not less than 98% of coumaphos, and its melting point is 95°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses and seeds

Weigh 10.0 g of the test sample, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for two hours.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of saturated sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of ethyl acetate/*n*-hexane (1:4) and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer ethyl acetate and *n*-hexane layers to a 300-ml conical flask. Add 50 ml of ethyl acetate/*n*-hexane (1:4) to the aqueous layer and repeat the above procedure, and then add ethyl acetate and *n*-hexane layers to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the ethyl acetate and *n*-hexane at 40°C or lower.

Add 30 ml of *n*-hexane to the residue and transfer the mixture to a 100-ml separating funnel, and then add 30 ml of *n*-hexane-saturated acetonitrile. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the acetonitrile layer to the rotary vacuum evaporator. Add 30 ml of *n*-hexane-saturated acetonitrile to the *n*-hexane layer and repeat the above procedure twice, and then combine the acetonitrile layer into the rotary vacuum evaporator. Remove the acetonitrile at 40°C or lower and dissolve the residue in 5-ml of acetone/*n*-hexane (1:1).

ii. Fruit, vegetables, teas and hops

In case of fruit and vegetables, weigh accurately 1 kg of the test sample and add an appropriate amount of water, if required. Homogenize the mixture and mesure out a sample equivalent to 20.0 g.

In case of teas, weigh 5.00 g of the test sample and add 20 ml of water, and then leave it to stand for two hours.

In case of hops, weigh 5.00 g of the test sample, previously ground, and add 20 ml of water, and then leave it to stand for two hours.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick

layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone and homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30ml at 40℃ or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of saturated sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of ethyl acetate/*n*-hexane (1:4) and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave the it to stand, and then transfer the ethyl acetate and *n*-hexane layers to a 300-ml conical flask. Add 50 ml of ethyl acetate/*n*-hexane (1:4) to the aqueous layer and repeat the above procedure twice, and then combine the ethyl acetate/*n*-hexane lyers to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper. Add the washings into the rotary vacuum evaporator and remove the ethyl acetate and *n*-hexane at 40°C or lower. Dissolve the residue in 5 ml of acetone/*n*-hexane (1:1).

iii. Foods except those listed in i and ii above

Obtains the extracts according to the methods described in i or ii.

b. Clean-up

Add 5 g of silica gel for column chromatography (63-200 μm in particle size) suspended in acetone/*n*-hexane (1:1) into a chromatograph tube (15 mm in inner diameter, 300 mm in length) and add over approximately 5 g of sodium sulfate (anhydrous) into the column. Pour acetone/*n*-hexane (1:1) into the column until only a small amount remains on the packing of the column and pour the solution obtained by the extraction described in 4-a into the column. Pour 100 ml of acetone/*n*-hexane (1:1) into the column and collect the eluate into a rotary vacuum evaporator, and then remove the acetone and *n*-hexane at 40°C or lower. Dissolve the residue in acetone to make exactly 5 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions 1

Column: A silicate glass capillary column (0.53 mm in inner diameter, 10-30 m in length) coated with methyl silicone for gas chromatography to a thickness of 1.5 µm

Column temperature: Hold the column temperature at 80°C for one minute, followed by an increase of 8°C every minute until reaching 250°C, and hold for five minutes.

Inlet temperature: 230°C

Detector: Operate at 280°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate to the optimal condition. Adjust the flows of air and hydrogen to the optimal conditions.

Testing conditions 2

Column: A silicate glass capillary column (0.32 mm in inner diameter, 10-30 m in length) coated with 50% trifluoro propyl methyl silicone for gas chromatography to a thickness of 0.25 µm

Column temperature: Hold the column temperature at 70°C for one minute, followed by an increase of 25°C every minute until reaching 125°C, after which increase the temperature by 10°C every minute until reaching 235°C, and hold for 12 minutes.

Inlet temperature: 230°C

Detector: Operate at 280°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate optimal condition. Adjust the flows of air and hydrogen to the optimal conditions.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the same conditions described in 5-a. Test results obtained must be the same as those obtained for the reference standard. Determine the quantity

using either the peak height or peak area method, if required.

(9) Chloramphenicol analytical method

1. Apparatus

A liquid chromatograph/mass spectrometer (LC/MS or LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives.***

Acetonitrile: Acetonitrile produced for liquid chromatography.

Divinylbenzene-*N*-vinylpyrrolidine copolymer cartridge column (60 mg): A polyethylene column of 12-13 mm in inner diameter packed with 60 mg of divinylbenzen-*N*-vinylpyrrolidone copolymer, or a column equivalent to the specified one in separation capability.

Divinylbenzene-*N*-vinylpyrrolidine copolymer cartridge column (200 mg): A polyethylene column of 12-13 mm in inner diameter packed with 200 mg of divinylbenzen-*N*-vinylpyrrolidone copolymer, or a column equivalent to the specified one in separation capability.

Water: Water produced for liquid chromatography.

3. Reference standard

Chloramphenicol: This product contains not less than 99% of chloramphenicol, and its decomposition point is 208°C.

4. Procedure

a. Extraction

i. Honey

Weigh 5.00 g of the test sample, previously homogenized, and dissolve it in 20 ml of water.

ii. Royal jelly

Weigh 1.00 g of the test sample, previously homogenized, add 60 ml of methanol/1% metaphosphoric acid (3:2), and homogenize the mixture again. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 2-mm thick layer of diatomaceous earth. Wash the residue on the filter paper with 15 ml of methanol/1% metaphosphoric acid (3:2) and filter the washings by suction. Combine the filtrates into the rotary vacuum evaporator and concentrate the mixture to 2 ml at 45°C or lower.

iii. Foods other than those given in sections i and ii

Weigh 5.00 g of the test sample, previously ground. For muscle, remove the fat layer as much as possible before grinding. Add 100 ml of methanol/1% metaphosphoric acid (3:2) and homogenize the mixture again. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 2-mm thick layer of diatomaceous earth. Wash the residue on the filter paper with 10 ml of methanol/1% metaphosphoric acid (3:2) and filter the washings by suction. Combine the filtrates into the rotary vacuum evaporator and cocentrate the mixture to 30 ml at 45ºC or lower.

b. Clean-up

i. Honey

Pour 5 ml of methanol and 5 ml of water into a divinylbenzene-*N*-vinylpyrrolidine copolymer cartridge column (60 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 5 ml of 20% (vol) methanol, and discard the effluent. Pour 6 ml of 60% (vol) methanol and collect the eluate into a rotary vacuum evaporator and remove the methanol and water at 45°C or lower. Dissolve the residue in 1.0 ml of acetonitrile/water (3:7), which is used as the sample solution.

ii. Royal jelly

Pour 10 ml of methanol and 10 ml of water into a divinylbenzene-*N*-vinylpyrrolidine copolymer cartridge column (200 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 4 ml of water and 4 ml of 5% (vol) methanol and discard the effluent. Pour 10 ml of 60% (vol) methanol and collect the eluate into a rotary vacuum evaporator, and remove the methanol and water at 45°C or lower. Dissolve the residue in 1.0 ml of acetonitrile/water (3:7), which is used as the sample solution.

iii. Foods other than those given in sections i and ii

Pour 5 ml of methanol and 5 ml of water into a divinylbenzene-*N*-vinylpyrrolidine copolymer cartridge column (60 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 10 ml of water, and discard the effluent. Pour 10 ml of methanol and collect the eluate into a rotary vacuum evaporator and remove the methanol at 40°C or lower.

Dissolve the residue in 1.0 ml of acetonitrile/water (3:7), which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilanized silica gel (2–5 μm in particle size).

Column: A stainless steel tube (2.0–6.0 mm in inner diameter, 100–250 mm in length).

Column temperature: 40°C

Mobile phase: Use acetonitrile/10 mmol/l ammonium formate (3:7). Adjust the flow rate so that chloramphenicol flows out in approximately 5 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

(10) Chlorpromazine analytical method

1. Apparatus

A liquid chromatograph/tandem mass spectrometer (LC/MS/MS).

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Strongly acidic cation exchanger cartridge column (500 mg): A polyethylene column of 8-9 mm in inner diameter packed with 500 mg of benzenesulfonyl propyl silane-bonded silica gel, or a column equivalent to the specified one in separation capability.

Water: Water produced for liquid chromatography.

Methanol: Methanol produced for liquid chromatography.

3. Reference standard

Chlorpromazine hydrochloride: This product contains not less than 98% of chlorpromazine hydrochloride, and its melting point is

194-196°C.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 25 ml of ethyl acetate and 1 ml of 4 mol/l potassium carbonate solution. Centrifuge the mixture at 3,000 rpm for five minutes And transfer the ethyl acetate layer into a rotary vacuum evaporator. Add 25 ml of ethyl acetate to the residue and repeat the above procedure and transfer the ethyl acetate layer into the rotary vacuum evaporator, and then remove the ethyl acetate at 40°C or lower. Add 30 ml each of acetonitrile and acetonitrile-saturated *n*-hexane to the residue and shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the acetonitrile layer to a 100-ml separating funnel. Add 30 ml of acetonitrile-saturated *n*-hexane and repeat the above procedure. Collect the acetonitrile layer into the rotary vacuum evaporator and remove the acetonitrile at 40°C or lower. Dissolve the residue in 10 ml of methanol/1.2% metaphosphoric acid solution (2:3) and filter the mixture through a cotton plug.

b. Clean-up

Pour 3 ml of methanol followed by 3 ml of water into a strongly acidic cation exchanger cartridge column (500 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 5 ml of water and discard the effluent. Pour 15 ml of methanol/0.1 mol/l dipotassium hydrogen orthophosphate solution (9:1) to the column and collect the eluate into a rotary vacuum evaporator, and then remove the water and methanol at 40°C or lower. Dissolve the residue in 1.0 ml of methanol, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5 μm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use acetonitrile/formic acid/water (500:1:500). Adjust the flow rate so that chlorpromazine flows out in approximately 15 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

(11) Diethylstilbestrol analytical method

1. Apparatus

A liquid chromatograph/tandem mass spectrometer (LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Glucuronidase solution: Should contain 100,000 units/ml of beta-D-glucuronidase extracted from *Helix pomatia* and refined.

Dichloromethane: Dichloromethane (special grade).

Weakly basic anion exchanger cartridge column (500 mg): A polyethylene column of 8-9 mm in inner diameter packed with 500 mg of diethylaminopropyl weakly basic anion exchanger produced for column chromatography, or a column equivalent to the specified one in separation capability.

Water: Water produced for liquid chromatography.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as dichloromethane before use..

3. Reference standard

Diethylstilbestrol: This product contains not less than 99% of diethylstilbestrol, and its decomposition point is 208°C.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 11 ml of 0.04 mol/l sodium acetate solution, and then add acetic acid to adjust the pH to 4.25-4.75. Add 100 µl of glucuronidase solution to the mixture and leave it to stand for 14 hours at 37°C. Add 16 ml of acetonitrile and shake the mixture vigorously for five minutes using a shaker. Centrifuge the mixture at 3,000 rpm for five minutes and transfer the supernatant into a rotary vacuum evaporator. Add 16 ml of acetonitrile to the residue and repeat the above procedure twice and transfer the supernatant into the rotary vacuum evaporator, and then concentrate the solution to approximately 3 ml at 50°C or lower. Add 50 ml of dichloromethane and 200 ml of 5% sodium chloride solution to the concentrated solution, and then shake it vigorously for five minutes using a shaker. Collect the dichloromethane layer and dehydrate with sodium sulfate (anhydrous). Add 50 ml of dichloromethane to the aqueous layer and repeat the above procedure twice, and then combine the dichloromethane layer. Add an adequate amount of sodium sulfate (anhydrous) and leave it to stand for 15 minutes with occasional shaking. Filter the content into a rotary vacuum evaporator and remove the dichloromethane at 40°C or lower. Dissolve the residue is in 4 ml of *n*-hexane/benzene (3:1).

b. Clean-up

Pour 10 ml of *n*-hexane/benzene (3:1) into a weakly basic anion exchanger cartridge column (500 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 2 ml of *n*-hexane/benzene (3:1) and 4 ml of dichloromethane, and discard the effluent. Pour 8 ml of dichloro methane/methanol (9:1) into the column and collect the eluate into a rotary vacuum evaporator, and then remove the dichloromethane and methanol at 40°C or lower. Dissolve the residue in 0.5 ml of acetonitrile/water (1:1), which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5 μm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use acetonitrile/0.002 mol/l ammonium acetate (1:1). Adjust the flow rate so that diethylstilbestrol flows out in approximately seven minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

(12) Analytical method for dimetridazole, metronidazole and ronidazole

1. Apparatus

A liquid chromatograph/tandem mass spectrometer (LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Methanol: Methanol produced for liquid chromatography.

Water: Water produced for liquid chromatography.

3. Reference standard

Dimetridazole: This product contains not less than 99% of dimetridazole, and its melting point is 138-139°C.

Metronidazole: This product contains not less than 99% of metronidazole, and its melting point is 158-160°C.

Ronidazole: This product contains not less than 99% of ronidazole, and its melting point is 167-169°C.

4. Procedure

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 20 ml of acetonitrile. Centrifuge the mixture at 3,000 rpm for five minutes and transfer the acetonitrile layer to a 100-ml separating funnel. Add 20 ml of *n*-hexane and shake the mixture vigorously, and then leave it to stand. Transfer the acetonitrile layer into a rotary vacuum evaporator and add 5 ml of *n*-propanol, and then remove the acetonitrile and *n*-propanol at 40°C or lower. Dissolve the residue in

1.0 ml of methanol and filter the mixture through a membrane filter with a pore size of 0.2 µm, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5 µm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use acetonitril/water (1:9). Adjust the flow rate so that dimetridazole, metronidazole and ronidazole flow out in 4-10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

(13) Daminozide analytical method

1. Apparatus

A gas chromatograph with an alkali flame ionization detector (GC-FID) or highly-sensitive nitrogen phosphorus detector (GC-NPD), a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS), and steam distillation apparatus. The steam distillation apparatus should be made of glass and roughly as shown in the following figure:

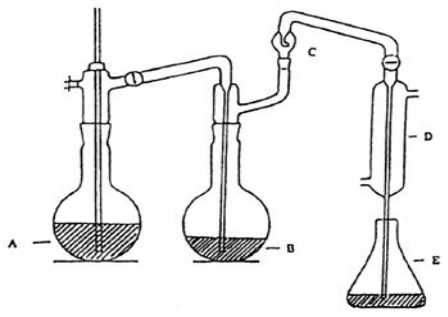
A: 1,000-ml round bottom flask (for steam generation)

B: 1,000-ml round bottom flask (for distillation)

C: Distillation trap

D: Cooling tube

E: 100-ml conical flask



2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those

listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Basic alumina cartridge column (1,710 mg): A polyethylene column of 8-9 mm in inner diameter packed with 1,710 mg of basic alumina, or a column equivalent to the specified one in separation capability.

Silicone for defoaming: Silicone produced for defoaming.

o-Nitrobenzaldehyde: *o*-Nitrobenzaldehyde (special grade).

1% *o*-Nitrobenzaldehyde-methanol solution: Dissolve *o*-nitrobenzaldehyde (100 mg) in 10 ml of methanol. Prepare immediately before use.

Phenolphthalein reagent: Dissolve phenolphthalein (1 g) in 100 ml of ethanol.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Methanol: Use a rotary vacuum evaporator on 300 mL of methanol to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Phosphate buffer solution (pH 5): Dissolve monopotassium dihydrogen monophosphate (13.15 g) and dipotassium hydrogen orthophosphate (0.59 g) in water to make a 100-ml solution.

3. Reference standard

Dimethylhydrazine: This product contains not less than 97% of

1,1-dimethylhydrazine, and its boiling point is 62-64°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses, seeds, fruit, vegetables, matcha and hops

In case cereal grains, legumes/pulses and seeds, weigh 5.0 g of test samples so as to pass through a standard mesh sieve (420 μm).

In case fruit and vegetables, weigh accurately 1 kg of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and measure out a sample equivalent to 10.0 g.

In case of matcha, weigh 5.0 g of the test sample.

In case of hops, weigh 5.0 g of the test sample, previously ground.

Then, add 80 ml of water to the obtained sample and shake it vigorously for 30 minutes, and then filter the mixture by suction through a glass fiber filter. Collect the residue on the filter and add 40 ml of water, and then shake the mixture for five minutes. Repeat the above procedure and combine the filtrate to a 1,000-ml round bottom flask (for distillation).

ii. Teas except matcha

Weigh 6.0 g of the test sample and soak it in 360 ml of water at 100°C, and then leave it to stand at room temperature for five minutes. Transfer 120 ml of the cooled filtrate into a 1,000-ml round bottom flask (for distillation).

iii. Foods except those listed in i and ii above

Obtain the extracts according to the methods described in i.

b. Distillation

Add 60 g of sodium hydroxide (65 g for vegetables and fruit) bit by bit into the round bottom flask described above with the flask water cooling. Add 1-2 drops of defoaming silicone to the dissolved solution and attach the flask to a distillation apparatus immediately. Separately, attach a 100-ml conical flask containing 5 ml of phosphate buffer solution (pH 5) and one drop of phenolphthalein reagent to a steam distiller and heat a 1,000-ml round bottom flask (for steam generation). Distill the solution until the distillate comes to 45 ml and confirm that the distillate remains colorless. Adjust the heater so as to complete the distillation in about 15 minutes.

c. Derivatization

Add 1 ml of 1% *o*-nitrobenzaldehyde-methanol solution to the above distillate and shake the mixture, and then leave it to stand for two hours at 30°C. Add 50 ml of *n*-hexane and shake the mixture for five minutes, and then leave it to stand. Collect the *n*-hexane layer and filter in a 200-ml eggplant-shaped flask through a liquid phase separation filter paper. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then combine the *n*-hexane layer to the eggplant-shaped flask above. Wash the residue on the filter with 10 ml of *n*-hexane and combine the washings to the eggplant-shaped flask above, and then remove the *n*-hexane at 40°C or lower. Dissolve the residue in 5 ml of acetone/*n*-hexane (1:19).

d. Clean-up

Pour 10 ml of acetone/*n*-hexane (1:19) into a basic alumina cartridge column (1,710 mg) and discard the effluent. Pour the solution obtained in 4-c into the column followed by 10 ml of acetone/ *n*-hexane (1:19) and collect the eluate into a rotary vacuum evaporator, and then remove the acetone and *n*-hexane at 40°C or lower. Dissolve the residue in acetone to make exactly 5 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard under the procedure described in 4-c.

Testing conditions

Column: A silicate glass capillary column (0.25 mm in inner diameter, 10-30 m in length) coated with 5% phenyl methyl silicone for gas chromatography to a thickness of 0.25 μm

Column temperature: Hold the column temperature at 60°C for two minutes, followed by an increase of 10°C every minute until reaching 280°C, and hold for five minutes.

Inlet temperature: 280°C

Detector: Operate at 280°C.

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate so that dimethylhydrazine derivatives flow out in approximately 13 minutes. Adjust the flows of air and hydrogen to the optimal

conditions.

b. Quantitative tests

Determine the content of dimethylhydrazine from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method. Furthermore, determine the content of daminozide using the following formula:

Daminozide content (ppm) = 2.67 x dimethylhydrazine content (ppm)

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the same conditions described in 5-a. Test results obtained for the reference standard must be the same as those obtained under the procedure described in 4-c. Determine the quantity using either the peak height or peak area method, if required.

d. Calibration curve

Assay a solution of dimethylhydrazine (1 ml) combined with 5 ml of phosphate buffer solution (pH 5) and 40 ml of water with the same procedure described in 4-c.

(14) Nitrofurans analytical method

Analyse 3-amino-2-oxazolidinone, 1-aminohydantoin, 3-amino-5-morpholinomethyl-2-oxazolidinone and semicarbazide.

1. Apparatus

A liquid chromatograph/tandem mass spectrometer (LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as “special grade” in this section must meet the requirements for “special grade” specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Porous diatomaceous earth column (to hold 20 ml of solution): A polyethylene column of 20-30 mm in inner diameter packed with granular porous diatomaceous earth produced for column chromatography that can hold 20 ml of solution, or a column equivalent to the specified one in separation capability.

o-Nitrobenzaldehyde: *o*-Nitrobenzaldehyde (special grade).

Water: Water produced for liquid chromatography.

3. Reference standard

3-amino-2-oxazolidinone: This product contains not less than 99% of 3-amino-2-oxazolidinone, and its decomposition point is 65-67°C.

1-aminohydantoin hydrochloride: This product contains not less than 90% of 1-aminohydantoin hydrochloride, and its decomposition point is 201-205°C.

3-amino-5-morpholinomethyl-2-oxazolidinone: This product contains not less than 99% of 3-amino-5-morpholinomethyl-2-oxazolidinone and its decomposition point is 115-120°C.

Semicarbazide hydrochloride: This product contains not less than 99% of semicarbazide hydrochloride, and its decomposition point is 175-177°C.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 70 ml of 0.1 mol/l hydrochloric acid. Centrifuge the mixture at 2,500 rpm for five minutes and collect the supernatant, and then add 0.1 mol/l hydrochloric acid to make 100 ml of solution.

b. Derivatization

Transfer 10 ml of the solution obtained by the extraction described in 4-a and add 0.4 ml of 0.05 mol/l *o*-nitrobenzaldehyde-dimethyl sulfoxide solution, and then leave it to stand for 16 hours at 37°C. Add 5 ml of 0.1 mol/l dipotassium hydrogen orthophosphate solution to the solution and add approximately 0.8 ml of a 1 mol/l sodium hydroxide solution to adjust the pH to 7-8. When some residue remains in the solution, centrifuge at 2,500 rpm for five minutes and collect the supernatant.

b. Clean-up

Pour the solution obtained by the derivatization described in 4-b into a porous diatomaceous earth column (to hold 20 ml of solution). Leave the column to stand for five minutes and add 100 ml of ethyl acetate.Collect the eluate into a rotary vacuum evaporator and remove the ethyl acetate at 40°C or lower. Dissolve the residue in 1.0 ml of acetonitrile/water (1:1), which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results

obtained must be the same as those obtained for the reference standards under the procedure described in 4-b and 4-c..

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5 μm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use a mixture of acetonitrile and 0.1% acetic acid solution. Create a concentration gradient of 1:4 to 4:1 acetonitrile/ 0.1% acetic acid solution in 15 minutes. Adjust the flow rate so that 3-amino-2-oxazolidinone flows out in approximately 12 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

(15) Propham analytical method

1. Apparatus

A gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as “special grade” in this section must meet the requirements for “special grade” specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is

found to include any substance that may interfere with the analysis of substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Octadecylsilane-bonded silica gel cartridge column (1,000 mg): A polyethylene column of 12-13 mm in inner diameter packed with 1,000 mg of octadecylsilane-bonded silica gel, or a column equivalent to the specified one in separation capability.

Graphite carbon and aminopropylsilane-bonded silica gel laminated cartridge column (500 mg/ 500 mg): A polyethylene column of 12-13 mm in inner diameter packed with 500 mg each of graphite carbon and aminopropylsilane-bonded silica gel, or a column equivalent to the specified one in separation capability.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

Toluene: Use a rotary vacuum evaporator on 300 mL of toluene to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

0.5 mol/l phosphate buffer solution (pH 7.0): Dissolve 136.85 g of secondary sodium phosphate (dodecahydrate) and 17.92 g of sodium phosphate monobasic (dihydrate) in water to make a 1,000-ml solution.

3. Reference standard

Propham: This product contains not less than 99% of propham, and its

melting point is 87°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses and seeds

Weigh 10.0 g of test samples, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for 15 minutes.

Then, add 50 ml of acetonitrile and homogenize the mixture for three minutes. Filter the homogenized sample by suction through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 20 ml of acetonitrile, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate, and then add acetonitrile to make a 100-ml solution.

Transfer 20 ml of the above solution to a 100-ml separating funnel and add 10 g of sodium chloride and 20 ml of 0.5 mol/l phosphate buffer solution (pH 7.0). Shake the mixture vigorously for 10 minutes using a shaker and leave it to stand, and then collect the acetonitrile layer.

Pour 10 ml of acetonitrile into an octadecylsilane-bonded silica gel cartridge column (1,000 mg) and discard the effluent. Pour the above acetonitrile layer into the column and collect the eluate in a 50-ml conical flask Pour 2 ml of acetonitrile into the column and combine the eluate to the flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 10 ml of acetonitrile and wash twice the residue on the surface of the filter paper with. the washings. Add the washings into the rotary vacuum evaporator and remove the acetonitrile at 40°C or lower. Dissolve the residue in 2 ml of toluene/acetonitril (1:3).

ii. Fruit, vegetables, teas and hops

In case of fruit and vegetables, weigh accurately 1 kg of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and measure out a sample equivalent to 20.0 g.

In case of teas and hops, weigh 5.00 g of the test sample, previously

ground, and add 20 ml of water, and then leave it to stand for 15 minutes.

Then, add 50 ml of acetonitrile and homogenize the mixture for three minutes. Filter the homogenized sample by suction through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 20 ml of acetonitrile, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate, and then add acetonitrile to make a 100-ml solution.

Transfer 20 ml of the above solution to a 100-ml separating funnel and add 10 g of sodium chloride and 20 ml of 0.5 mol/l phosphate buffer solution (pH 7.0). Shake the mixture vigorously for 10 minutes using a shaker and leave it to stand, and then collect the acetonitrile layer in a 50-ml conical flask. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 10 ml of acetonitrile and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the acetonitrile at 40°C or lower. Dissolve the residue in 2 ml of toluene/acetonitril (1:3).

iii. Foods except those listed in i and ii above

Obtain the extracts according to the methods described in i or ii.

b. Clean-up

Pour 10 ml of toluene/acetonitrile (1:3) into a graphite carbon and aminopropylsilane-bonded silica gel laminated cartridge column (500 mg/500 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 20 ml of toluene/acetonitrile (1:3). Collect the eluate into a rotary vacuum evaporator and remove the toluene and acetonitrile at 40°C or lower. Dissolve the residue in acetone/*n*-hexane (1:1) to make exactly 2 ml (1 ml for cereal grains, legumes/pulses, seeds, teas and hops) of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference

standard.

Testing conditions

Column: A silicate glass capillary column (0.25 mm in inner diameter, 30 m in length) coated with 5% phenyl methyl silicone for gas chromatography to a thickness of 0.25 μm

Column temperature: Hold the column temperature at 50°C for one minute, followed by an increase of 25°C every minute until reaching 125°C, after which increase the temperature by 10°C every minute until reaching 300°C, and hold for 6.5 minutes.

Inlet temperature: 250°C

Detector: Operate at 280°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate to the optimal conditions.

Injection method: Splitless injection method.

Measured mass size (m/z): 93, 137 and 179

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

(16) Malachite green analytical method

Analyze malachite green and leucomalachite green.

1. Apparatus

A liquid chromatograph/mass spectrometer (LC/MS or LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Tolutions, Etc.***, Part II ***Food Additives***. Reagents designated as “special grade” in this section must meet the requirements for “special grade” specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Ammonium formate: Ammonium formate (special grade).

Strongly acidic cation exchanger cartridge column (500 mg): A polyethylene column of 8-9 mm in inner diameter packed with 500 mg of benzenesulfonyl propylsilanized silica gel, or a column equivalent to the specified one in separation capability.

Citric acid–phosphate buffer (pH 3.0)

Solution 1: Dissolve 63.0g of citric acid in water to make 1,000 mL of

solution.

Solution 2: Dissolve 215 g of disodium phosphate in water to make 1,000 mL of solution.

Mix Solution 1 and Solution 2 and adjust the pH to 3.0.

Dichloromethane: Dichloromethane (special grade).

Water: Water produced for liquid chromatography.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). When it contains substances that may interfere with the analysis, wash it with ethyl acetate before using.

3. Reference standard

Malachite Green Oxalate: This product contains not less than 99% of malachite green oxalate, and its decomposition point is 164ºC.

Leucomalachite Green: This product contains not less than 99% of leucomalachite green, and its decomposition point is 103ºC.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 10 mL of citric acid-phosphate buffer (pH 3.0). Add 15 mL of acetonitrile and shake the mixture vigorously for five minutes using a shaker. Centrifuge the mixture at 3,000 rpm for five minutes and collect the acetonitrile-water layer. Add 15mL of acetonitrile to the residue and repeat the above procedure, and then combine the acetonitrile layer with the acetonitrile-water layer.

Add 5 mL of *n*-hexane to the combined layers and shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then collect the acetonitrile-water layer. Add 5 mL of *n*-hexane to the collected layer and repeat the above procedure to collect the acetonitrile-water layer.

Add 50 mL of 20% sodium chloride and 10 mL of dichloromethane to the collected layer and shake it vigorously for five minutes using the shaker and leave it to stand, and then collect the acetonitrile-dichloromethane layer.

Add an adequate amount of sodium sulfate (anhydrous) and leave it to stand for 15 minutes with occasional shaking, and then filter the mixture.

b. Clean-up

Pour 5 mL of acetonitrile into a strongly acidic cation exchanger

cartridge column (500 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4–a into the column followed by 5 mL of acetonitrile and discard the effluent. Pour 10 mL of acetonitrile/ammonia water (9:1) and collect the eluate into a rotary vacuum evaporator, and then remove the acetonitrile and ammonia water at 40ºC or lower. Dissolve the residue in 1.0 mL of acetonitrile, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standards.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5μm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use a mixture of acetonitrile and 0.01 mol/L ammonium formate. Create a concentration gradient of 1:9 to 1:0 acetonitrile/0.01 mol/L ammonium formate in 20 minutes, and maintain a ratio of 1:0 for 10 minutes. Adjust the flow rate so that malachite green flows out in approximately 10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the same conditions described in 5-a using either the peak height or peak area method.

6. Regardless of the provisions in 5, ingredients of agricultural chemicals and other chemical substances listed in the first column of the table in (1) must not be contained in foods at levels exceeding the limits stipulated in the third column of the same table according to the food categories shown in the second column of the same table. In association with this regulation, foods listed in the “foods” column in the table in (2) below shall be tested using the part listed in the “samples” column in the table as a sample. In addition, in the substances used as ingredients of agricultural chemicals and other chemical substances listed in the first

column of the table in (1) below, which are stipulated to be "Not detected" in the third column of the same table in the foods listed in the second column of the same table, no ingredient of agricultural chemicals or other chemical substances shall be detected when these foods are tested using the test methods stipulated in (3) to (10) below.

(1) The maximum residue limits of substances used as ingredients of agricultural chemicals in foods (***MRLs List***)

(2) Samples

Food	Sample material
Barley and buckwheat	Threshed seeds
Wheat and rye	Husked seeds
Rice (brown rice)	Husked seeds
Corn (maize)	Seeds with the husk, the silk and the cores removed
Other cereal grains	Threshed seeds
Peas, beans (dry)*, broad beans and soybeans (dry)	Seeds without the pods
Peanuts, dry	With the shells removed
Other legumes/pulses	Seeds without the pods
Apricot, mume plum, cherry, Japanese plum (including prune) and nectarine	With the peduncles and the seeds removed
Peach	With the skins and the seeds removed
Orange (including navel orange), grapefruit, citrus *natsudaidai* (whole) , lime and lemon	Whole fruit
Citrus *natsudaidai* (pulp) and *unshu* orange (pulp)	With the peels removed
Citrus *natsudaidai*, peels	With the calyxes removed
Other citrus fruits	Whole fruit
Pear, Japanese pear, quince and apple	With the blossom scars, the cores and the peduncles removed
Loquat	With the peduncles, the skins and the seeds removed
Avocado and mango	With the seeds removed

Kiwifruit	With the skins removed
Guava	With the calyxes removed
Date	With the calyxes and the seeds removed
Pineapple	With the tops removed
Passion fruit and papaya	Whole fruit
Banana	With the pedicels removed
Strawberry, cranberry, huckleberry, blackberry and blueberry	With the calyxes removed
Raspberry	Whole fruit
Other berries	With the calyxes removed
Japanese persimmon	With the calyxes and the seeds removed
Watermelon, *makuwauri* melon and melons	With the rinds removed
Grape	With the peduncles removed
Other fruits	Edible portions
Turnip (roots) and Japanese radish (roots, including radish)	With the dirt lightly rinsed off with water
Turnip (leaves), watercress, kale, Japanese radish (leaves, including radish) and brussels sprouts	With the decayed leaves removed
Cauliflower and broccoli	With the leaves removed
Cabbage and Chinese cabbage	A sample consisting of one portion from each of four heads, with each head equally cut into four portions, without the decayed outer leaves and the cores.
Kyona and *komatsuna* (Japanese mustard spinach)	With the roots and the decayed leaves removed
Horseradish	Roots with the dirt lightly rinsed off with water
Qing-geng-cai and other cruciferous vegetables	Edible portions
Sweet potato, konjac, taro, potato, yam and other potatoes	With the dirt lightly rinsed off with water

Pumpkin (including squash), cucumber (including gherkin) and oriental pickling melon (vegetable)	With the vines removed
Other cucurbitaceous vegetables	Edible portions
Artichoke, endive and chicory	With the decayed leaves removed
Burdock and salsify	A sample thinly sliced then ground with a meat grinder, the leaves having been removed and the dirt having been lightly rinsed off with water
Shungiku	With the roots and the decayed leaves removed
Lettuce (including cos lettuce and leaf lettuce)	With the decayed outer leaves and the cores removed
Other composite vegetables	Edible portions
Shiitake mushroom, button mushroom and other mushrooms	Edible portions
Celery, parsley and *mitsuba*	With the roots and the decayed leaves removed
Carrot and parsnip	With the dirt lightly rinsed off
Other umbelliferous vegetables	Edible portions
Tomato, egg plant and pimiento (sweet pepper)	With the calyxes removed
Other solanceous vegetables	Edible portions
Asparagus	Stems
Onion, garlic, welsh (including leek) and multiplying onion	With the outer skins and the root hair removed
Nira and other liliaceous vegetables	Edible portions
Green soybeans, kidney beans (with pods, immature) and peas (with pods, immature)	With the pedicels removed
Okra	With the calyxes removed
Sugarcane	With the husks removed
Ginger	With the leaves removed, and with the dirt lightly rinsed off with water

Sugar beet	With the dirt lightly rinsed off with water
Spinach	With red roots left on, and with the root hair and the decayed leaves removed
Bamboo shoots and other vegetables	Edible portions
Sesame seeds, rapeseeds, sunflower seeds, safflower seeds, cotton seed and other oil seeds	Seeds only
Almond, ginkgo nut, chestnut, walnut, pecan and other nuts	With the husks removed
Cacao beans and coffee beans	Beans without the pods
Tea	Tea leaves
Hop	Dried flowers
Other spices and other herbs	Edible portions

(3) 2,4,5-T analytical method

Perform according to 5 (3).

(4) Amitrol analytical method

Perform according to 5 (5).

(5) Analytical method for aldrin, endrin and dieldrin

1. Apparatus

A gas chromatograph with an electron capture detector (GC-ECD) and a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to

concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Ether: Use a rotary vacuum evaporator on 300 mL of diethyl ether to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is found to include any substance that may interfere with the analysis of substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Synthetic magnesium silicate (Florisil) for column chromatography: Heat florisil (150-250 μm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

3. Reference standard

Aldrin: This product contains not less than 97% of aldrin, and its melting point is 102-104°C.

Endrin: This product contains not less than 98% of endrin, and its decomposition point is 200°C.

Dieldrin: This product contains not less than 98% of dieldrin, and its melting point is 177-179°C.

4. Procedure

a. Extraction

i. Cereal grains, legumes/pulses and seeds

Weigh 10.0 g of test samples, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for two hours.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then add the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washing into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower.

Add 20 ml of *n*-hexane to the residue and transfer the mixture to a 100-ml separating funnel, and then add 40 ml of *n*-hexane-saturated acetonitrile. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the acetonitrile layer into the rotary vacuum evaporator. Add 40 ml of *n*-hexane-saturated acetonitrile to the *n*-hexane layer and repeat the above procedure twice, and then combine the acetonitrile layer into the rotary vacuum evaporator. Remove the acetonitrile at 40°C or lower and

dissolve the residue in *n*-hexane to make exactly 5 ml of solution.

ii. Fruit, vegetables, matcha

In case of fruit and vegetables, weigh accurately 1 kg of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and measure out a sample equivalent to 20.0 g.

In case of matcha, weigh 5.00 g of the test sample and add 20 ml of water, and then leave it to stand for two hours.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of 10% sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of *n*-hexane and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 50 ml of *n*-hexane to the aqueous layer and repeat the above procedure twice, and then combine the *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower. Dissolve the residue in *n*-hexane to make exactly 10 ml of solution.

iii. Teas (limited to unfermented tea) other than matcha

Weigh 9.00 g of the test sample and soak it in 540 ml of water at 100°C, and the leave it to stand at room temperature for five minutes. Transfer 360 ml of the cooled filtrate into a 500-ml conical flask. Add 100 ml of acetone and 2 ml of saturated lead acetate

solution to the above flask and leave it to stand for one hour at room temperature. Filter the mixtue by suction into a 1,000-ml separating funnel through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Wash the above conical flask with 50 ml of acetone and wash the residue on the surface of the filter paper with the washings. Add the washings into the above separating funnel and also add 30 g of sodium chloride and 100 ml of *n*-hexane. Shake the mixture vigorously for five minutes and leave it to stand, and then transfer the *n*-hexane layer to a 300-ml conical flask. Add 100 ml of *n*-hexane to the aqueous layer and repeat the above procedure, and then add the *n*-hexane layer to the conical flask above. Add an adequate amount of asodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washings into the rotary vacuum evaporator and remove the *n*-hexane at 40°C or lower. Dissolve the residue in *n*-hexane to make exactly 5 ml of solution.

b. Clean-up

Add 10 g of florisil for column chromatography suspended in *n*-hexane into a chromatograph tube (15 mm in inner diameter, 300 mm in length) and add over approximately 5 g of sodium sulfate (anhydrous) into the column. Spill out the *n*-hexane until only a small amount remains on the packing of the column and pour 2 ml of the solution obtained by the extraction described in 4-a into the column. Pour 200 ml of ether/*n*-hexane (3:17) into the column and collect the eluate into a rotary vacuum evaporator, and then remove the ether and *n*-hexane at 40°C or lower. Dissolve the residue in *n*-hexane to make exactly 2 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions 1

Column: A silicate glass capillary column (0.25 mm in inner diameter, 10-30 m in length) coated with methyl silicone for gas

chromatography to a thickness of 0.25 μm.

Column temperature: Hold the column temperature at 50°C for one minute, followed by an increase of 25°C every minute until reaching 175°C, after which increase the temperature by 10°C every minute until reaching 300°C, and hold for five minutes.

Inlet temperature: 230°C

Detector: Operate at 300°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate so that aldrin flows out in approximately 10 minutes.

Testing conditions 2

Column: A silicate glass capillary column (0.25 mm in inner diameter, 10-30 m in length) coated with 14% cyanopropylphenyl-methyl silicone for gas chromatography to a thickness of 0.25 μm..

Column temperature: Hold the column temperature at 80°C for two minutes, followed by an increase of 30°C every minute until reaching 190°C, after which increase the temperature by 3.6°C every minute until reaching 250°C, and hold for eight minutes.

Inlet temperature: 230°C

Detector: Operate at 300°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate so that aldrin flows out in approximately 10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the conditions described in 5-a. Test results obtained must be the same as those obtained for the reference standard. Determine the quantity using either the peak height or peak area method, if required.

(6) Captafol analytical method

Perform according to 5 (6).

(7) Quinoxaline-2-carboxylic acid analytical method

Performed according to 5 (7).

(8) Cyhexatin analytical method

Performed according to 5 (4).

(9) Daminozide analytical method

Performed according to 5 (13).

(10) Analytical method for triazophos and parathion

1. Apparatus

A gas chromatograph with an alkali flame ionization detector (GC-FID), a flame photometric detector (GC-FPD, interference filter for phosphorus determination, wavelength: 526 nm), or a highly-sensitive nitrogen phosphorus detector (GC-NPD), and a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as "special grade" in this section must meet the requirements for "special grade" specified in the Japan Industrial Standards for the reagents.

Acetonitrile: Use a rotary vacuum evaporator on 300 mL of acetonitrile to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 µL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Acetone: Use a rotary vacuum evaporator on 300 mL of acetone to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 µL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Sodium chloride: Sodium chloride (special grade). If the reagent is found to include any substance that may interfere with the analysis of substances to be analyzed from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Silica gel for column chromatography (63-200 µm in particle size): Heat silica gel made for column chromatography (63-200 µm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Diatomaceous earth: Diatomaceous earth for chemical analysis.

Ethyl acetate: Use a rotary vacuum evaporator on 300 mL of ethyl acetate to concentrate to the point of dryness. Dissolve the residue in 5 mL of *n*-hexane. For analysis, inject 5 µL of the solution into a

GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2 x 10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

Sodium sulfate (anhydrous): Sodium sulfate (anhydrous)(special grade). If the reagent is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

3. Reference standard

Triazophos: This product contains not less than 98% of triazophos, and its melting point is 0-5°C.

Parathion: This product contains not less than 97% of parathion, and its boiling point is 375°C.

4. Procedure

a. Extraction

i. Cereal grains and legumes/pulses

Weigh 10.0 g of test sample, previously ground so as to pass through a standard mesh sieve (420 μm). Add 20 ml of water to the obtained sample and leave it to stand for two hours.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator through a filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixtue to approximately 30ml at 40℃ or lower.

Transfer the concentrated solution to a 300-ml separating funnel already containing 100 ml of saturated sodium chloride solution.

Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of ethyl acetate/*n*-hexane (1:4) and add the washings to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the layers of ethyl acetate and *n*-hexane to a 300-ml conical flask. Add 50 ml of ethyl acetate/*n*-hexane (1:4) to the aqueous layer and repeat the above procedure, and then add the layers of ethyl acetate and *n*-hexane to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper with the washings. Add the washing into the rotary vacuum evaporator and remove the ethyl acetate and *n*-hexane at 40°C or lower.

Add 30 ml of *n*-hexane the residue and transfer the mixture to a 100-ml separating funnel, and then add 30 ml of *n*-hexane-saturated acetonitrile. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the acetonitrile layer into the rotary vacuum evaporator. Add 30 ml of *n*-hexane-saturated acetonitrile to the *n*-hexane layer and repeat the above procedure twice, and then combine the acetonitrile layer into the rotary vacuum evaporator above. Remove the acetonitrile at 40°C or lower and dissolve the residue in 5 ml of acetone/*n*-hexane (1:1).

ii. Fruit and vegetables

Weigh accurately 1 kg of the test sample and add an appropriate amount of water to the sample, if required. Homogenize the mixture and measure out a sample equivalent to 20.0 g.

Then, add 100 ml of acetone and homogenize the mixture for three minutes. Filter the homogenized sample by suction into a rotary vacuum evaporator using filter paper covered with a 1-cm thick layer of diatomaceous earth. Collect the residue on the surface of the filter paper and add 50 ml of acetone, and then homogenize the mixture for three minutes. Repeat the above procedure and combine the filtrate into the rotary vacuum evaporator, and then concentrate the mixture to approximately 30 ml at 40°C or lower.

Transfer the concentrated solution to a 300-ml separating funnel

already containing 100 ml of saturated sodium chloride solution. Wash the eggplant-shaped flask of the above rotary vacuum evaporator with 100 ml of ethyl acetate/*n*-hexane (1:4) and add the washing to the separating funnel above. Shake the mixture vigorously for five minutes using a shaker and leave it to stand, and then transfer the layers of ethyl acetate and *n*-hexane to a 300-ml conical flask. Add 50 ml of ethyl acetate/*n*-hexane (1:4) to the aqueous layer and repeat the above procedure twice, and then combine the ethyl acetate and *n*-hexane layer to the conical flask above. Add an adequate amount of sodium sulfate (anhydrous) to the flask and leave it to stand for 15 minutes with occasional shaking. Filter the content of the flask into a rotary vacuum evaporator. Wash the flask with 20 ml of *n*-hexane and wash twice the residue on the surface of the filter paper. Add the washings into the rotary vacuum evaporator and remove the ethyl acetate and *n*-hexane at 40°C or lower. Dissolve the residue in 5 ml of acetone/*n*-hexane (1:1).

b. Clean-up

Add 5 g of silica gel for column chromatography (63-200 µm in particle size) suspended in acetone/*n*-hexane (1:1) into a chromatograph tube (15 mm in inner diameter, 300 mm in length) and add over approximately 5 g of sodium sulfate (anhydrous) to the column. Pour acetone and *n*-hexane (1:1) into the column until only a small amount remains on the packing of the column. Pour the solution obtained by the extraction described in 4-a into the column followed by 100 ml of acetone/*n*-hexane (1:1) and collect the eluate into a rotary vacuum evaporator, and then remove the acetone and *n*-hexane at 40°C or lower. Dissolve the residue in acetone to make exactly 5 ml of solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions 1

Column: A silicate glass capillary column (0.53 mm in inner diameter, 10-30 m in length) coated with methyl silicone for gas chromatography to a thickness of 1.5 µm.

Column temperature: Hold the column temperature at 80°C for one minute, followed by an increase of 8°C every minute until reaching 250°C, and hold for five minutes.

Inlet temperature: 230°C

Detector: Operate at 280°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate to the optimal condition. Adjust the flows of air and hydrogen to the optimal conditions.

Testing conditions 2

Column: A silicate glass capillary column (0.32 mm in inner diameter, 10-30 m in length) coated with 50% trifluoro propyl methyl silicone for gas chromatography to a thickness of 0.25 μm.

Column temperature: Hold the column temperature at 70°C for one minute, followed by an increase of 25°C every minute until reaching 125°C, after which increase the temperature by 10°C every minute until reaching 235°C, and hold for 12 minutes.

Inlet temperature: 230°C

Detector: Operate at 280°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate to the optimal condition. Adjust the flows of air and hydrogen to the optimal conditions.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the conditions described in 5-a. Test results obtained must be the same as those obtained for the reference standard. Determine the quantity using either the peak height or peak area method, if required.

7. In addition to the substances stipulated under 6, ingredients of agricultural chemicals and other chemical substances listed in the first column of the table in (1) must not be contained in foods at levels exceeding the limits stipulated in the third column of the same table according to the food categories shown in the second column of the same table. Concerning this, foods listed in the “foods” column in the table in

(2) shall be tested using the part listed in the “samples” column in the table as a sample. In addition, in the substances used as ingredients of agricultural chemicals and other chemical substances listed in the first column of the table in (1), which are stipulated to be “Not detected” in the third column of the same table in the foods listed in the second column of the same table, no ingredient of agricultural chemicals or other chemical substances shall be detected when these foods are tested using the test methods stipulated in (3) to (8).”

(1) The maximum residue limits of substances used as ingredients of agricultural chemicals in foods (***Provisional MRLs List***)

(2) Samples

Food	Sample material
Barley and buckwheat	Threshed seeds
Wheat and rye	Husked seeds
Rice (brown rice)	Husked seeds
Corn (maize)	Seeds with the husk, the silk and the cores removed
Other cereal grains	Threshed seeds
Peas, beans (dry)*, broad beans and soybeans (dry)	Seeds without the pods
Peanuts, dry	With the shells removed
Other legumes/pulses	Seeds without the pods
Apricot, mume plum, cherry, Japanese plum (including prune) and nectarine	With the peduncles and the seeds removed
Peach	With the skins and the seeds removed
Orange (including navel orange), grapefruit, citrus *natsudaidai* (whole) , lime and lemon	Whole fruit
Citrus *natsudaidai* (pulp) and *unshu* orange (pulp)	With the peels removed
Citrus *natsudaidai*, peels	With the calyxes removed
Other citrus fruits	Whole fruit

Pear, Japanese pear, quince and apple	With the blossom scars, the cores and the peduncles removed
Loquat	With the peduncles, the skins and the seeds removed
Avocado and mango	With the seeds removed
Kiwifruit	With the skins removed
Guava	With the calyxes removed
Date	With the calyxes and the seeds removed
Pineapple	With the tops removed
Passion fruit and papaya	Whole fruit
Banana	With the pedicels removed
Strawberry, cranberry, huckleberry, blackberry and blueberry	With the calyxes removed
Raspberry	Whole fruit
Other berries	With the calyxes removed
Japanese persimmon	With the calyxes and the seeds removed
Watermelon, *makuwauri* melon and melons	With the rinds removed
Grape	With the peduncles removed
Other fruits	Edible portions
Turnip (roots) and Japanese radish (roots, including radish)	With the dirt lightly rinsed off with water
Turnip (leaves), watercress, kale, Japanese radish (leaves, including radish) and brussels sprouts	With the decayed leaves removed
Cauliflower and broccoli	With the leaves removed
Cabbage and Chinese cabbage	A sample consisting of one portion from each of four heads, with each head equally cut into four portions, without the decayed outer leaves and the cores
Kyona and *komatsuna* (Japanese mustard spinach)	With the roots and the decayed leaves removed

Horseradish	Roots with the dirt lightly rinsed off with water
Qing-geng-cai and other cruciferous vegetables	Edible portions
Sweet potato, konjac, taro, potato, yam and other potatoes	With the dirt lightly rinsed off with water
Pumpkin (including squash), cucumber (including gherkin) and oriental pickling melon (vegetable)	With the vines removed
Other cucurbitaceous vegetables	Edible portions
Artichoke, endive and chicory	With the decayed leaves removed
Burdock and salsify	A sample thinly sliced then ground with a meat grinder, the leaves having been removed and the dirt having been lightly rinsed off with water
Shungiku	With the roots and the decayed leaves removed
Lettuce (including cos lettuce and leaf lettuce)	With the decayed outer leaves and the cores removed
Other composite vegetables	Edible portions
Shiitake mushroom, button mushroom and other mushrooms	Edible portions
Celery, parsley and *mitsuba*	With the roots and the decayed leaves removed
Carrot and parsnip	With the dirt lightly rinsed off
Other umbelliferous vegetables	Edible portions
Tomato, egg plant and pimiento (sweet pepper)	With the calyxes removed
Other solanceous vegetables	Edible portions
Asparagus	Stems
Onion, garlic, welsh (including leek) and multiplying onion	With the outer skins and the root hair removed
Nira and other liliaceous vegetables	Edible portions

Green soybeans, kidney beans (with pods, immature) and peas (with pods, immature)	With the pedicels removed
Okra	With the calyxes removed
Sugarcane	With the husks removed
Ginger	With the leaves removed, and with the dirt lightly rinsed off with water
Sugar beet	With the dirt lightly rinsed off with water
Spinach	With red roots left on, and with the root hair and the decayed leaves removed
Bamboo shoots and other vegetables	Edible portions
Sesame seeds, rapeseeds, sunflower seeds, safflower seeds, cotton seed and other oil seeds	Seeds only
Almond, ginkgo nut, chestnut, walnut, pecan and other nuts	With the husks removed
Cacao beans and coffee beans	Beans without the pods
Tea	Tea leaves
Hop	Dried flowers
Other spices and other herbs	Edible portions

(3) Analytical method for aldrin, endrin and dieldrin

Perform according to 6 (5).

(4) Clenbuterol analytical method

1. Apparatus

A liquid chromatograph/tandem mass spectrometer (LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Strongly acidic cation exchanger cartridge column (500 mg): A polyethylene column of 8-9 mm in inner diameter packed with 500 mg of benzenesulfonil propyl silane-bonded silica gel, or a column equivalent to the specified one separation capability.

Water: Water produced for liquid chromatography.

Methanol: Methanol produced for liquid chromatography.

3. Reference standard

Clenbuterol hydrochloride: This product contains not less than 98% of clenbuterol hydrochloride, and its melting point is 174-176°C.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 25 ml of ethyl acetate and 1 ml of 4 mol/l potassium carbonate solution. Centrifuge the mixture at 3,000 rpm for five minutes and collect the ethyl acetate layer into a rotary vacuum evaporator. Add 25 ml of ethyl acetate to the residue and repeat the above procedure and combine the ethyl acetate layer, and then remove the ethyl acetate at 40°C or lower. Add 30 ml each of acetonitrile and acetonitrile-saturated *n*-hexane to the residue and shake the mixture vigorously for five minutes using a shaker, and then leave it to stand. Transfer the acetonitrile layer to a 100-ml separating funnel and add 30 ml of acetonitrile-saturated *n*-hexane. Repeat the above procedure and collect the acetonitrile layer into the rotary vacuum evaporator, and then remove the acetonitrile at 40°C or lower. Dissolve the residue in 10 ml of methanol/1.2% metaphosphoric acid solution (2:3) and filter the mixture through a cotton plug.

b. Clean-up

Pour 3 ml of methanol followed by 3 ml of water into a strongly acidic cation exchanger cartridge column (500 mg) and discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 5 ml of water and discard the effluent. Pour 15 ml of methanol/0.1 mol/l dipotassium hydrogen ortho phosphate solution (9:1) into the column and collect the eluate into a rotary vacuum evaporator, and then remove the water and methanol at 40°C or lower. Dissolve the residue in 1.0 ml of methanol, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5 μm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use acetonitrile/formic acid/water (500:1:500). Adjust the flow rate so that clenbuterol flows out in approximately 10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

(5) Dexamethasone analytical method

1. Apparatus

A liquid chromatograph/tandem mass spectrometer (LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***.

Acetonitrile: Acetonitrile produced for liquid chromatography.

Octadecylsilane-bonded silica gel cartridge column (360 mg): A polyethylene column of 8-9 mm in inner diameter packed with 360 mg of octadecylsilane-bonded silica gel, or a column equivalent to the specified one in separation capability.

Synthetic magnesium silicate (Florisil) for column chromatography: Heat florisil (150-250 μm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Water: Water produced for liquid chromatography.

Methanol: Methanol produced for liquid chromatography.

Phosphate buffer solution (pH 5.0):

Solution 1: Dissolve 27.2 g of monopotassium dihydrogen mono phosphate in water to make a 1,000 ml of solution.

Solution 2: Dissolve 3.48 g of dipotassium hydrogen orthophosphate in water to make a 100 ml of solution.

Mix Solution 1 and Solution 2 and adjust the pH to 5.0.

3. Reference standard

Dexamethasone: This product contains not less than 99% of dexamethasone, and its melting point is 262-264°C.

4. Procedure

a. Extraction

Weigh 5.00 g of the test sample, previously ground, and homogenize it with 30 ml of 95% acetonitrile solution. Centrifuge the mixture at 2,500 rpm for five minutes and collect the acetonitrile layer. Add 30 ml of 95% acetonitrile solution to the residue and repeat the above procedure, and then collect the acetonitrile layer.

b. Clean-up

i. Florisil for column chromatography

Add 8 g of florisil for column chromatography suspended in acetonitrile into a chromatograph tube (15 mm in inner diameter, 300 mm in length). Spill out the acetonitrile until only a small amount remains on the packing of the column and pour 100 ml of acetonitrile, and then discard the effluent. Pour the solution obtained by the extraction described in 4-a into the column followed by 30 ml of acetonitrile and then collect the eluate into a 300-ml separating funnel. Add 50 ml of *n*-hexane the funnel and shake it vigorously using a shaker for three minutes, and then leave it to stand. Collect the acetonitrile layer into a rotary vacuum evaporator and remove the acetonitrile at 40°C or lower. Dissolve the residue in 4 ml of phosphate buffer solution (pH 5.0) and add 6 ml of water.

ii. Octadecylsilane-bonded silica gel column chromatography

Pour 10 ml of methanol, 10 ml of water and 2 ml of phosphate buffer solution (pH 5.0) into the octadecylsilane-bonded silica gel cartridge column (360 mg) in that order, and discard the effluent. Pour the solution obtained by chromatography described in 4-b-i into the column followed by 5 ml of phosphate buffer solution (pH 5.0) and 10 ml of 25% methanol solution,.and then discard the effluent. Pour 10 ml of 60% acetonitrile solution into the column and collect the eluate into a rotary vacuum evaporator, and then remove the acetonitrile and water at 40°C or lower. Dissolve the

residue in 0.5 ml of 10% acetonitrile solution, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (2-5 μm in particle size).

Column: A stainless tube (2.0-6.0 mm in inner diameter, 100-250 mm in length).

Column temperature: 40°C

Mobile phase: Use acetonitrile/formic acid/water (1,200:1:800). Adjust the flow rate so that dexamethasone flows out in approximately 7-10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

(6) Analytical method for triazophos and parathion

Perform according to 6 (10).

(7) Analytical method for alpha-trenbolone and beta-trenbolone

1. Apparatus

A high-performance liquid chromatograph with an ultraviolet spectrophotometric detector (HPLC-UV) and a liquid chromatograph/ mass spectrometer (LC/MS or LC/MS/MS)

2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***. Reagents designated as “special grade” in this section must meet the requirements for “special grade” specified in the Japan Industrial Standards for the reagents.

Weakly basic anion exchanger cartridge column (500 mg): A polyethylene column of 8-9 mm in inner diameter packed with 500 mg of diethylaminopropyl weakly basic anion exchanger produced for

column chromatography, or a column equivalent to the specified one in separation capability.

Hydroxypropyl dextran for column chromatography: Dextran (25-100 μm in particle size) chemically bonded with a hydroxypropyl group produced for column chromatography.

Dichloromethane: Dichloromethane (special grade).

3. Reference standard

Alpha-trenbolone: This product contains not less than 94% of alpha-trenbolone, and its melting point is 110°C.

Beta-trenbolone: This product contains not less than 99% of beta-trenbolone, and its melting point is 186°C.

4. Procedure

a. Extraction

Weigh 5.00 g out of test sample, previously ground. For muscle, remove the fat layer as much as possible before grinding.

Add 20 ml of acetonitrile/methanol (4:1) and homogenize the mixture again. Centrifuge at 2,600 rpm for five minutes and collect the supernatant in a 100-ml separating funnel. Add 20 ml of acetonitrile/methanol (4:1) to the precipitate and centrifuge the mixture under the above conditions, and then combine the acetonitrile and methanol layer to the separating funnel above. Add 20 ml of water-saturated *n*-hexane to the funnel and shake it vigorously for five minutes, and then leave it to stand. Transfer the acetonitrile and methanol layer to a 200-ml separating funnel. Add 40 ml each of 5% sodium sulfate solution and dichloromethane to the funnel and shake it vigorously for five minutes and leave it to stand, and then transfer the dichloromethane layer into a rotary vacuum evaporator. Add 15 ml of dichloromethane to the aqueous layer and repeat the above procedure and combine the dichloromethane layer into the rotary vacuum evaporator, and then remove the dichloromethane at 40°C or lower. Dissolve the residue in 1 ml of *n*-hexane/benzene (3:1).

b. Clean-up

i. Weakly basic anion exchanger column chromatography

Pour 6 ml of *n*-hexane/benzene (3:1) into a weakly basic anion exchanger cartridge column (500 mg) and discard the effluent. Pour the the solution obtained by the extraction described in 4-a into the column followed by 2 ml of *n*-hexane/benzene (3:1) and discard the

effluent. Pour 3 ml of dichloromethane/methanol (9:1) into the column and collect the eluate into a rotary vacuum evaporator, and then evaporate the solvent to near dryness under nitrogen at 40°C or lower. Dissolve the residue in 0.5 ml of benzene/methanol (17:3).

ii. Hydroxypropyl dextran for column chromatography

Add hydroxypropyl dextran for column chromatography suspended in benzene/methanol (17:3), previously left to stand for 12 hours, into a polyethylene column (6 mm in inner diameter) so as to obtain a 120-mm long layer of dextran. Spill out the benzene/methanol (17:3) until only a small amount remains on the packing of the column. Pour the solution obtained by chromatography described in 4-b-i into this column followed by 10 ml of benzene/methanol (17:3). Discard 2.0 ml of the first effluent and collect the next eluate into a rotary vacuum evaporator, and then evaporate the solvent to near dryness under nitrogen at 40°C or lower. Dissolve the residue in 0.5 ml of acetonitrile/water (1:1), which is used as the sample solution.

5. Determination

a. Qualitative tests

Perfom qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column packing: Octadecylsilane-bonded silica gel (5 µm in particle size).

Column: A stainless tube (4.0-6.0 mm in inner diameter, 150 mm in length).

Column temperature: 40°C

Detector: Operate with an absorption wavelength of 340 nm.

Mobile phase: Use acetonitrile/water (5:6). Adjust the flow rate so that alpha-trenbolone flows out in approximately 10 minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform liquid chromatography/mass spectrometry under the

conditions described in 5-a. Test results obtained must be the same as those obtained for the reference standard. Determine the quantity using either the peak height or peak area method, if required.

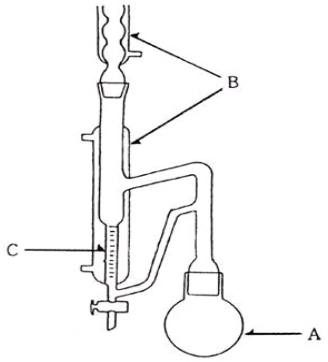
(8) Ethylene dibromide analytical method

1. Apparatus

A gas chromatograph with an electron capture detector (GC-ECD), a gas chromatograph/mass spectrometer (GC/MS or GC/MS/MS) and Dean Stark distillation apparatus

The Dean Stark distillation apparatus is roughly as shown in the following figure:

A: Distillation flask
B: Cooling tube
C: Distillation trap



2. Reagents and test solutions

In addition to the reagents and test solutions listed below, use those listed in Section C ***Reagents/Test Solutions, Etc.***, Part II ***Food additives***.

Synthetic magnesium silicate (Florisil) for column chromatography: Heat florisil (150-250 μm in particle size) at 130°C for 12 hours or longer. Cool down to room temperature in a desiccator.

Silicone for defoaming: Silicone produced for defoaming.

n-Hexane: Use a rotary vacuum evaporator on 300 mL of *n*-hexane to evaporate until 5 mL is left. For analysis, inject 5 μL of the solution into a GC-ECD. Peaks other than that of *n*-hexane in the resulting chromatogram should be as high as or lower than the peaks of γ-BHC at 2×10^{-11}g.

Water: Distilled water. If the distilled water is found to include any substance that may interfere with analysis of the target compositional substances from agricultural chemicals, wash with a solvent such as *n*-hexane before use.

3. Reference standard

Ethylene dibromide: This product contains not less than 99% of

ethylene dibromide, and its boiling point is 131.5°C.

4. Procedure

a. Extraction

Homogenize about 1 kg of a test sample and transfer 100 g of the homogenized sample into a 1,000-ml distillation flask, and then add 200 ml of water and 10 ml of *n*-hexane to the flask. Add a few drops of defoaming silicone and boiling stones to the mixture and attach the flask to the Dean Stark distillation apparatus to be heated and refluxed for one hour. After cooling, remove most of the water in the distillation trap and filter the *n*-hexane layer into a 10-ml measuring flask through a liquid phase separation filter paper. Wash the trap with a small amount of *n*-hexane and filter the washings through the filter paper above. Combine the filtrate with the *n*-hexane layer to make exactly a 10-ml solution.

b. Clean-up

Transfer the solution obtained by the extraction described in 4-a to a 10-ml test tube with a glass stopper. Add approximately 1 g of florisil for column chromatography to the test tube and shake it vigorously, and then leave it to stand at room temperature for about 15 minutes. Collect about 5 ml of supernatant in another test tube with a glass stopper, which is used as the sample solution.

5. Determination

a. Qualitative tests

Perform qualitative tests under the following conditions. Test results obtained must be the same as those obtained for the reference standard.

Testing conditions

Column: A silicate glass capillary column (0.32 mm in inner diameter, length in 30 m) coated with 6% cyanopropylphenyl-methyl silicone for gas chromatography to a thickness of 1.8 µm.

Column temperature: Hold the column temperature at 50°C for two minute, followed by an increase of 5°C every minute until reaching 110°C, after which increase the temperature by 30°C every minute until reaching 260°C, and hold for five minutes.

Inlet temperature: 250°C

Detector: Operate at 300°C

Gas flow rate: Use helium as the carrier gas. Adjust the flow rate so

that ethylene dibromide flows out in approximately eight minutes.

b. Quantitative tests

Determine the quantity from the test results obtained under the conditions described in 5-a using either the peak height or peak area method.

c. Confirmation tests

Perform gas chromatography/mass spectrometry under the conditions described in 5-a. Test results obtained must be the same as those obtained for the reference standard. Determine the quantity using either the peak height or peak area method, if required.

8. In cases where a substance used as an ingredient of an agricultural chemical (excluding substances determined by the Minister of Health, Labour and Welfare as having no potential to cause damage to human health under Paragraph 3 of Article 11 of the Law), for which no compositional standards are stipulated in 5 to 7, is the same as the substance naturally contained in the food, the amount of the substance contained in the food shall not exceed the amount normally contained in the food. This, however, does not apply to foods containing substances that may cause damage to human health at the amount normally contained.

9. In cases where substances used as ingredients of agricultural chemicals and other chemical substances that are listed in the first column of the following table are contained in a food, the amount of the substances shall not exceed the amount stipulated in the third column of the same table according to the food categories shown in the second column of the same table.

The maximum residue limits of substances used as ingredients of agricultural chemicals in foods (excluding foods listed in the second column of the table in 6 (1) and the second column of the table in 7 (1)):

Column 1	Column 2	Column 3
AZINPHOS-METHYL	Other spices, dried	0.5 ppm
ACEPHATE	Other spices, dried (except the fruit of *Sansho* (Japanese pepper))	0.2 ppm

AMITRAZ	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the Japanese Agricultural Standards (JAS) for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
ALDICARB	Peanut oil (limited to refined peanut oil and peanut salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.01 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.01 ppm
IPRODIONE	Other spices, dried (limited to seeds)	0.05 ppm
	Other spices, dried (limited to roots or rhizomes)	0.1 ppm
IMIDACLOPRID	Wheat flour (except whole grain)	0.03 ppm
	Wheat bran	0.3 ppm
ETHION	Other spices, dried (limited to fruits)	5 ppm
	Other spices, dried (limited to seeds)	3 ppm
	Other spices, dried (limited to roots or rhizomes)	0.3 ppm
ETHEPHON	Raisin	5 ppm
	Fig, dried	10 ppm

ENDOSULFAN	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
	Other spices, dried (limited to fruits)	5 ppm
	Other spices, dried (limited to seeds)	1 ppm
	Other spices, dried (limited to roots or rhizomes)	0.5 ppm
CARBARYL	Rice bran	170 ppm
	Milled rice	1 ppm
	Wheat flour (except whole grain)	0.2 ppm
	Wheat germ	1 ppm
	Wheat bran	2 ppm
	Corn oil (except edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
	Soybean oil (except edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm
	Tomato juice	3 ppm
	Tomato paste	10 ppm
	Edible olive oil (limited to virgin oil)	25 ppm
	Sunflower oil (except edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm

QUINTOZENE	Other spices, dried (limited to fruits)	0.02 ppm
	Other spices, dried (limited to seeds)	0.1 ppm
	Other spices, dried (limited to roots or rhizomes)	2 ppm
GLYPHOSATE	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
GLUFOSINATE	Sunflower oil (except edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Rapeseed oil (except refined rapeseed oil and rapeseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
KRESOXIM-METHYL	Raisin	2 ppm
	Edible olive oil (limited to virgin olive oil)	0.7 ppm
CLETHODIM	Soybean oil (limited to edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm

	Soybean oil (except edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	1 ppm
	Sunflower oil (except edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
	Cottonseed oil (limited to refined cottonseed oil and cotton seed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
	Cottonseed oil (except refined cottonseed oil and cotton seed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
	Rapeseed oil (limited to refined rapeseed oil and rapeseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
	Rapeseed oil (except refined rapeseed oil and rapeseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
CHLORDANE	Milled rice	0.02 ppm

	Soybean oil (limited to edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.02 ppm
	Soybean oil (except edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Linseed oil (except refined linseed oil)	0.05 ppm
CHLORPYRIFOS	Wheat flour (except whole grain)	0.1 ppm
	Corn oil (limited to edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm
	Soybean oil (limited to edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm

	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Other spices, dried (limited to fruits)	1 ppm
	Other spices, dried (limited to seeds)	5 ppm
	Other spices, dried (limited to roots or rhizomes)	1 ppm
CHLORPYRIFOS-METHYL	Wheat flour (except whole grain)	2 ppm
	Wheat bran	20 ppm
	Other spices, dried (limited to fruits)	0.3 ppm
	Other spices, dried (limited to seeds)	1 ppm
	Other spices, dried (limited to roots or rhizomes)	5 ppm
CHLORMEQUAT	Wheat flour (limited to whole grain)	5 ppm
	Wheat flour (except whole grain)	2 ppm
	Wheat bran	10 ppm
	Rye flour (limited to whole grain)	4 ppm
	Rye flour (except whole grain)	3 ppm
	Rye bran	10 ppm
	Rapeseed oil (except refined rapeseed oil and rapeseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
FENBUTATIN OXIDE	Plum, dried	10 ppm
	Raisin	20 ppm

DICHLORVOS and NALED (as total)	Wheat flour (limited to whole grain)	2 ppm
	Wheat flour (except whole grain)	1 ppm
	Wheat bran	10 ppm
	Wheat germ	10 ppm
	Other spices, dried	0.1 ppm
DIQUAT	Milled rice	0.2 ppm
	Wheat flour (limited to whole grain)	2 ppm
	Wheat flour (except whole grain)	0.5 ppm
	Wheat bran	5 ppm
	Vegetable oil (except refined vegetable oil)	0.05 ppm
DICOFOL	Plum, dried	3 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
	Other spices, dried (limited to fruits)	0.1 ppm
	Other spices, dried (limited to seeds)	0.05 ppm
	Other spices, dried (limited to roots or rhizomes)	0.1 ppm
DISULFOTON	Other spices, dried	0.05 ppm

CYHALOTHRIN	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.02 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.02 ppm
DIPHENYLAMINE	Apple juice	0.5 ppm
CYPRODINIL	Wheat bran	2 ppm
	Plum, dried	5 ppm
	Raisin	5 ppm
CYPERMETHRIN	Other spices, dried (limited to fruits)	0.1 ppm
	Other spices, dried (limited to roots or rhizomes)	0.2 ppm
	Vegetable oil (limited to refined vegetable oil)	0.5 ppm
DIMETHIPIN	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
DIMETHOATE	Edible olive oil (except virgin oil)	0.05 ppm
	Other spices, dried (limited to fruits)	0.5 ppm

	Other spices, dried (limited to seeds)	5 ppm
	Other spices, dried (limited to roots or rhizomes)	0.1 ppm
BROMIDE	Wheat flour (limited to whole grain)	50 ppm
	Peach, dried	50 ppm
	Plum, dried	20 ppm
	Raisin	100 ppm
	Date, dried	100 ppm
	Fig, dried	250 ppm
	Fruits (dried fruits except peach, plum, grape, date and fig)	30 ppm
	Other herbs, dried	400 ppm
SPINOSAD	Wheat bran	2 ppm
	Raisin	1 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.01 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.01 ppm
DIAZINON	Plum, dried	2 ppm
	Other spices, dried (limited to fruits)	0.1 ppm
	Other spices, dried (limited to seeds)	5 ppm
	Other spices, dried (limited to roots or rhizomes)	0.5 ppm

THIODICARB and METHOMYL	Wheat flour (except whole grain)	0.03 ppm
	Wheat germ	2 ppm
	Wheat bran	3 ppm
	Corn oil (limited to edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.02 ppm
	Soybean oil (limited to edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm
	Soybean oil (except edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.04 ppm
TEBUCONAZOLE	Raisin	3 ppm
TEBUFENOZIDE	Raisin	2 ppm
DELTAMETHRIN and TRALOMETHRIN (as total)	Wheat flour (limited to whole grain)	2 ppm
	Wheat flour (except whole grain)	0.3 ppm
	Wheat bran	5 ppm
TERBUFOS	Rapeseed oil (except refined rapeseed oil and rapeseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm

PARAQUAT	Sunflower oil (limited to edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Sunflower oil (except edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
PARATHION	Other spices, dried (limited to fruits)	0.2 ppm
	Other spices, dried (limited to seeds)	0.1 ppm
	Other spices, dried (limited to roots or rhizomes)	0.2 ppm
PARATHION-METHYL	Raisin	1 ppm
	Other spices, dried (limited to fruits)	5 ppm
	Other spices, dried (limited to seeds)	5 ppm
	Other spices, dried (limited to roots or rhizomes)	3 ppm
BIORESMETHRIN	Wheat flour (limited to whole grain)	1 ppm
	Wheat flour (except whole grain)	1 ppm
	Wheat germ	3 ppm
	Wheat bran	5 ppm
BIFENTHRIN	Wheat flour (limited to whole grain)	0.5 ppm
	Wheat flour (except whole grain)	0.2 ppm

	Wheat bran	2 ppm
PIPERONYL BUTOXIDE	Wheat flour (limited to whole grain)	30 ppm
	Wheat flour (except whole grain)	10 ppm
	Wheat germ	90 ppm
	Wheat bran	80 ppm
	Corn oil (except edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	80 ppm
	Tomato juice	0.3 ppm
	Juice of citrus fruits	0.05 ppm
	Fruits, dried	0.2 ppm
PYRIPROXYFEN	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.01 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.01 ppm
PIRIMICARB	Other spices, dried (limited to seeds)	5 ppm
PIRIMIPHOS-METHYL	Wheat bran	15 ppm
	Other spices, dried (limited to fruits)	0.5 ppm
	Other spices, dried (limited to seeds)	3 ppm
PYRETHRINS	Fruits, dried	0.2 ppm
VINCLOZOLIN	Other spices, dried	0.05 ppm

FAMOXADONE	Wheat bran	0.2 ppm
	Raisin	5 ppm
FENAMIPHOS	Peanut oil (except refined peanut oil and peanut salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
FENITROTHION	Wheat flour (limited to whole grain)	5 ppm
	Wheat bran	20 ppm
	Other spices, dried (limited to fruits)	1 ppm
	Other spices, dried (limited to seeds)	7 ppm
	Other spices, dried (limited to roots or rhizomes)	0.1 ppm
FENTHION	Edible olive oil (limited to virgin oil)	1 ppm
PHENTHOATE	Other spices, dried (limited to seeds)	7 ppm
FENVALERATE	Wheat flour (limited to whole grain)	2 ppm
	Wheat flour (except whole grain)	0.2 ppm
	Wheat bran	5 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm

	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
FENPROPATHRIN	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	3 ppm
FLUDIOXONIL	*Nira*, dried Basil, dried	50 ppm 50 ppm
FLUSILAZOLE	Raisin	1 ppm
FLUTOLANIL	Rice bran Milled rice	10 ppm 1 ppm
PROCHLORAZ	Wheat bran Sunflower oil (limited to edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	7 ppm 1 ppm
PROCYMIDONE	Sunflower oil (limited to edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
PROPARGITE	Corn flour Corn oil (limited to edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm 0.5 ppm

	Corn oil (except edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.7 ppm
	Peanut oil (limited to refined peanut oil and peanut salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.3 ppm
	Peanut oil (except refined peanut oil and peanut salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.3 ppm
	Orange juice	0.3 ppm
	Apple juice	0.2 ppm
	Grape juice	1 ppm
	Raisin	12 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm
PROFENOFOS	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
HEPTACHLOR	Soybean oil (limited to edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.02 ppm

	Soybean oil (except edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.5 ppm
PERMETHRIN	Wheat flour (limited to whole grain)	2 ppm
	Wheat flour (except whole grain)	0.5 ppm
	Wheat germ	2 ppm
	Wheat bran	5 ppm
	Soybean oil (except edible soybean oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
	Sunflower oil (limited to edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	1 ppm
	Sunflower oil (except edible sunflower oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	1 ppm
	Cottonseed oil (limited to refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.1 ppm
	Other spices, dried	0.05 ppm
PENCONAZOLE	Raisin	0.5 ppm
PHOSALONE	Other spices, dried (limited to fruits)	2 ppm
	Other spices, dried (limited to seeds)	2 ppm

	Other spices, dried (limited to roots or rhizomes)	3 ppm
PHORATE	Peanut oil (limited to refined peanut oil and peanut salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Peanut oil (except refined peanut oil and peanut salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.05 ppm
	Other spices, dried (limited to fruits)	0.1 ppm
	Other spices, dried (limited to seeds)	0.5 ppm
	Other spices, dried (limited to roots or rhizomes)	0.1 ppm
MALATHION	Tomato juice	0.01 ppm
	Other spices, dried (limited to fruits)	1 ppm
	Other spices, dried (limited to seeds)	2 ppm
	Other spices, dried (limited to roots or rhizomes)	0.5 ppm
METHAMIDOPHOS	Other spices, dried (except the fruit of *Sansho* (Japanese pepper))	0.1 ppm
METALAXYL and MEFENOXAM	Other spices, dried (limited to seeds)	5 ppm
METHIDATHION	Edible olive oil (limited to virgin oil)	2 ppm
	Cottonseed oil (except refined cottonseed oil and cottonseed salad oil that meet the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	2 ppm

METHOXYFENOZIDE	Plum, dried	2 ppm
	Raisin	3 ppm
METHOPRENE	Wheat flour (limited to whole grain)	5 ppm
	Wheat flour (except whole grain)	2 ppm
	Wheat bran	10 ppm
	Corn oil (limited to edible corn oil that meets the JAS for Edible Vegetable Fats and Oils, and other edible oils that meet standards equivalent to or stricter than JAS)	0.2 ppm
HYDROGEN PHOSPHIDE	Vegetables, dried	0.01 ppm
	Fruits, dried	0.01 ppm

10. In addition to the provisions of 6 or 9, when food products are manufactured or processed using foods for which compositional standards are specified in 6 through 9, the foods used must comply with the standards given in 6 through 9.