European and Mediterranean Plant Protection Organization Organisation Européenne et Méditerranéenne pour la Protection des Plantes

Efficacy evaluation of plant protection products Evaluation biologique des produits phytosanitaires

PP 1/242 (2) Taint tests

Specific scope

This Standard provides general guidance on the requirements for testing whether harvested plants or plant products are tainted by plant protection products. It explains the circumstances under which taint tests are necessary, how extensive they should be, where to obtain samples for the tests, how to collect and handle them and how to have them evaluated by tasting assessors. This Standard does not apply to plant products which are so transformed that they are totally different in nature from the raw crop (e.g. bread, beer, wine), which are covered by EPPO Standards PP 1/243 *Effects of plant protection products on transformation processes* and PP 1/268 *Study of unintentional effects of plant protection products on fermentation processes and characteristics of wine*.

Background

For certain types of treatments with plant protection products, it may be necessary to provide evidence that the use of the product does not give a taint (unpleasant taste or smell) to the harvested or processed plant product. A large number of factors can influence whether a product causes taint including the crop, climate, soil type, method of application, the interval from application to harvest and the method of processing. Due to the impracticality of investigating all of these, only factors which have been shown to be important are examined. These procedures will demonstrate whether the food product from a crop treated with a plant protection product is different in flavour from a control made from an untreated crop. In most cases, it is likely that no difference in taste will be found and the result may be taken to show the absence of taint from treatment. Where some difference is demonstrated, it may be possible to assess the taint on the basis of descriptions given by assessors. For definitions of this and other terms, see BSI (1992) or ISO (1992).

Historically, taint testing has often been targeted almost entirely at crops which subsequently undergo commercial processing, and most commonly those undergoing heating processes or quick freezing. This is because of the potential

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of such processes to concentrate or enhance any tainting effect. Information suggests that fungicides are the most likely group of products to cause taint, with plant growth regulators and sprout suppressants the least likely. Certain products have a high propensity to cause taint, and a high occurrence of taint is likely if they are used near to harvest or as post-harvest treatments. However, in general, applications near the harvest interval are not necessarily more likely to cause taint. For nematicides and insecticides, even certain soil-applied treatments at or before planting, have been associated with the occurrence of taint.

There is always a possibility of taint, even in freshly harvested produce, but the frequency of occurrence of taint on fresh produce is generally so low, and the burden of testing so enormous, that it is not practical to require routine taint testing of fresh produce. Taint tests on fresh produce are only advisable in cases where a specific risk exists.

Use of taint tests

There are no simple rules or cut-off criteria, to decide whether or not taint tests should be conducted, but the following generalizations may be made:

• The length of time between application and harvest has little impact on the propensity for taint;

• If an active substance, a product or a similar type of product has caused taint in one crop, it will have a higher potential for taint in other crops.

On the basis of these general principles, and of information on the historical occurrence of taint, risk situations can be classified as follows:

- High risk: taint test is normally required for: a product used on crops for processing, and/or a product containing an active substance known to have caused taint, or chemically similar to one known to cause taint, and/or a higharoma compound applied close to harvest;
- Medium risk: a taint test is useful in the case of: the presence of residues at harvest, and/or an active substance about which little is known, and/or a systemic compound applied to foliage;
- Low risk: a taint test is not usually required for: a product used on fresh produce only, and/or an active substance not associated with taint problems or relatively

similar in structure to active substances not associated with taint problems, and/or a product leaving no residues at harvest and/or a non-systemic compound not applied to harvestable plant parts.

If it is decided that taint testing is necessary, guidance is provided in Tables 1 and 2 on the principal crops which might initially be tested, the main processing methods for these crops, and the possibility of extrapolation to other crops. If tests have already been undertaken for a given plant protection product on several crops, and taint has not been detected, it should be possible to reduce the number of tests required or to discontinue testing. In particular, if no effects were observed with crops which are usually sensitive to taint, no further crops need to be tested.

If it is decided that taint testing is not necessary, arguments in support of this decision should be provided (e.g. mode of action of the active substance, method of uptake in the plant, long time interval to harvest, use of directed

 Table 1 Acceptable extrapolations between crops for taint

Crop group	Extrapolation from	Extrapolation to
Legumes	Vining pea PIBSX Dwarf bean PHSVN	Broad bean VICFX Broad bean VICFX, Runner bean PHSCO, Mange-tout
	Dried nea PIRSA	pea or sugar pea PIBSZ
Brassicas	Diled pea Tibb/t	
(excluding root brassicas)		
a) Head and leafy brassicas	Brussels sprouts BRSOF	Kale BRSOA, cabbage BRSOL/BRSOR
	Kale BRSOA	Brussels sprouts BRSOF, cabbage BRSOL/BRSOR
	Cabbage BRSOL/BRSOR	Kale BRSOA, Brussels sprouts BRSOF
b) Flowering brassicas	Cauliflower BRSOB	Broccoli BRSOK
	Broccoli BRSOK	Cauliflower BRSOB, kohlrabi BRSOG
Leafy vegetables	Spinach SPQOL	French sorrel RUMSC
	Lettuce LACSA	Endive CICEN/CICEC, leaf beet BEAVV
Root crops	Carrot DAUCS	Parsnip PAVSA
	Swede BRSNA	Turnip BRSRR
	Turnip BRSRR	Swede BRSNA
	Red beet BEAVD	_
Bulb vegetables	Bulb onions ALLCE	Garlic ALLSA, leek ALLPO
Stem vegetables	Celery APUGV	Celeriac APUGR
Solanaceous crops	Potato SOLTU	_
	Tomato LYPES	_
	Aubergine SOLME	_
Cucurbits	Cucumber CUMSA	Courgette/squash CUUPE, melon CUMME, watermelon CITLA
Soft fruit	Strawberry FRAAN	-
	Raspberry RUBID	Loganberry RUBLO, blackberry RUBFR, other <i>Rubus</i> spp. RUBSS
	Red/white currant RIBRU	Black currant RIBNI, gooseberry RIBUC
Pome fruit	Apple MABSD	Pear PYUCO, quince CYDOB
Stone fruit	Any <i>Prunus</i> PRNSS, except almond	Any other <i>Prunus</i> PRNSS, except almond
	Almond PRNDU	Walnut IUGRE, chestnut CSNSA
Mushrooms	Any mushroom	Any other mushroom

Crop group	Crop	Primary processing method	Secondary processing method*
Legumes	Vining pea	Freezing	Heat preservation
	Dwarf French bean	Freezing	(Heat preservation)
	Dried pea	Heat preservation	
Brassicas	Brussels sprout	Freezing	
	Cabbage	Freezing, fermentation	
	Cauliflower	Freezing	
	Calabrese/broccoli	Freezing	
Leafy vegetables	Spinach	Freezing	(Heat preservation)
Root crops	Carrot	Freezing	(Heat preservation)
	Swede	Freezing	(Heat preservation)
	Turnip	Freezing	(Heat preservation)
	Red beet	Heat preservation	
Bulb vegetables	Bulb onion	Freezing	
Potatoes	Potato	Freezing	Heat preservation
Soft fruit	Strawberry	Heat preservation	(Freezing)
	Raspberry	Heat preservation	(Freezing)
	Black currant	Heat preservation	(Freezing)
Top fruit	Apple	Heat preservation	(Freezing)
	Plum	Heat preservation	
Mushroom	Mushroom	Heat preservation	(Freezing)
Stem vegetables	Leek	Freezing	
	Celery	Freezing	

Table 2 Primary and secondary processing methods for crops

Freezing = quick freezing; Heat preservation = canning, preserving (jam) or juicing as appropriate. Other processing methods may also be considered.

*Where these are indicated in brackets, there is no necessity for testing, but the applicant may choose to test for his own purposes.

sprays, no contact with crop foliage, no root uptake). If no taint data is provided for evaluation by the registration authority, and a high or medium risk of taint is considered possible, then it may be appropriate to give advice on the product label, for example 'Consult processor before using on crops for processing'.

Field trials and chemical application

Field trial design and site

Specific trials may be set up for taint purposes alone. Trials design, recording and management should then comply with the principles laid down in national guidance for residue trials or by EPPO Standard PP 1/181 *Conduct and reporting of efficacy evaluation trials, including good experimental practice*, for efficacy trials. However, residue and efficacy trials may also be used for taint testing purposes and these trials should similarly comply with the appropriate guidance indicated above. The cultivars chosen should be representative of those used commercially for processing. The system of cultivation, picking, transport and storage etc., should be uniform for any one trial.

Test crops should be grown under a range of soil and climatic condition, in areas representative of the commercial crops. Due consideration should be given to the fitness of the harvested produce for processing and tasting (Appendix 1). To avoid deterioration of harvested produce, the place of testing and the time period from harvest to testing should be considered when deciding where to grow the crop.

Results from taint testing trials conducted in other areas or regions where registration is sought may also be taken into consideration, provided that agronomic, cultural and climatic conditions are broadly comparable between those regions. A justification of their relevance should normally be made.

The test methods given in Appendix 2 require equal quantities of control and treated material, i.e. the amount of control crop should be at least equal to the total of all treatments. In designing trials, account should be taken of the requirements for taint and other intended purposes of the trial (e.g. residues or efficacy) to ensure that there is sufficient material available to allow representative sampling, and that requirements for the two purposes are compatible.

If the previous cropping and treatment history of the trial area is known, any anomalous results due to residues from earlier treatments can be investigated. Records should thus be kept of all treatments, including fertilizers, so that the source of any interactions can be traced.

Testing facility or organization

The testing facility or organization which performs the tests should be identified and it should be clearly stated whether the trials are performed by an official or officially recognized testing facility or organization (see EPPO Standard PP 1/181 Conduct and reporting of efficacy evaluation trials, including good experimental practice). Tests done by official or officially recognized organizations are valid studies for registration authorities as well for processors.

Test product, dose, time and method of application

Applications should be made as stated on the product label or with the maximum dose, the maximum number of treatments and the latest time of application. Taint may be caused not only by the active substance but also by the formulants used in the plant protection product. Therefore, tests with the active substance alone are not acceptable. The test product should be the same formulation, as the product that is submitted for registration.

Table 1 should be used if it has been determined that taint testing is necessary, to determine the crops that may initially be tested for taint, and the crops to which extrapolation is acceptable. The generation of the appropriate number of acceptable results for the crop/s given in a cell in column two should allow extrapolation to the crop/s given in the adjacent cell in column three. The list of crops is given as an example; the table can be extended to other crops important in EPPO countries.

The method of application, and water volumes used, should be appropriate to the use of the product and as recommended on the product label. Where the label recommends use of the product with an adjuvant, e.g. wetting agent, this should be included in the treatment. If a product is recommended for application after harvest, then the interval between treatment and preparation of the crop should comply with the harvest interval on the label. If good commercial practice may result in a longer period of storage prior to processing, then account may also need to be taken of this. For seed treatments where taint testing is required, the interval between treatment and sowing should be recorded.

Sampling, handling and storage of the crop

Detailed guidance is given in Appendix 1.

Tasting tests

Detailed guidance on taint testing is given in Appendix 2. Any required authorization should be obtained before any treated product is consumed.

Guidance on the extent of taint testing required

Formulation

It may not be practical to test all formulations of one active substance. Therefore, the main commercial formulation

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should normally be tested. Where there is a significant change of formulation, and new data on residues is required, or where there are significant differences between formulations, additional testing may be required if this change is likely to lead to taint.

If a product submitted for registration is a new formulation of a well-known active substance that has never been associated with taint, it may be argued that further taint testing is unnecessary, e.g. change from a wettable granule (WG) formulation to a capsule suspension (CS) formulation.

Crops tested

Testing should be conducted initially on the main crops and cultivars on which the plant protection product is to be used. The extent of investigation on other crops will depend on their similarity to the main crop already tested, the quantity and quality of the data already available and how far the manner of use of the product and methods of processing the crops are similar. Guidance on acceptable extrapolations is given in Table 1.

Food uses tested

Taint tests should normally be undertaken using the primary processing methods for the crop. Check tests may also be undertaken to cover secondary methods of processing. Guidance on the primary and secondary processing methods for different crops is given in Table 2. Where both the primary and secondary methods proposed have been tested, it will not normally be necessary to cover further processing methods.

Number of tests

Normally trials should be conducted over 2 years, with the number of trials split equally between the 2 years as shown in Table 3. Where positive results are obtained from the primary processing method, or some doubt exists, further testing may be necessary. In such situations, two additional tests will generally be required, which may be undertaken in Year 2 or if necessary in a third year of testing. If results from the secondary processing methods show problems, then a full programme of testing as for primary processing may be required. If a specific taint is detected, various courses of action remain open. For instance, the applicant may prefer to label the product appropriately rather than to generate additional data.

The main formulation of a product may therefore be tested four times, normally over 2 years, using the primary processing method. In some instances, it may be possible to take samples for secondary processing from the same trials as for the primary processing methods (e.g. where growing conditions and cultivars are the same for both processing methods).

Table 3	The	number	of	tests	normally	required	for	taint	testing	per
main cro	р									

	Number of tests per main crop		
	Year 1	Year 2	
Primary processing method*	2	2	
Secondary processing* methods (check tests)	1	1	

*A guide to the primary and secondary processing methods for the major crops is given in Table 2.

If the results in year 1 show an absence of taint for products considered to present a high risk, the number of tests in year 2 may be reduced. If the results in year 1 show an absence of taint for products considered to present a low or medium risk, testing in year 2 would not be needed. Absence of taint can be assumed tentatively if all results are negative (not significant at P = 0.05) in the first year. Negative results over the full test period indicate absence of taint for the particular crop under the conditions tested. If some results are positive or some doubt exists, for example a small but not significant proportion of assessors detecting a taint, further testing may be necessary (see Appendix 2).

Only those taints which a reasonable proportion of the population can recognize can be detected by the proposed testing scheme. As the number of assessors used by the scheme is limited, and people's sensitivity to specific compounds may vary widely, it may happen that a taint which could only be recognized by sensitive subjects may not be detected. Scrutiny of replicate results for consistent correct identification of taint by a particular individual may give an indication of the presence of 'minority' taint, even though this was not detected in the overall results.

References

- ASTM (1996) *Manual 26 Sensory Testing Methods*, 2nd edn. American Society for Testing and Materials. http://www.astm.org. [accessed on 1 June 2014].
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- ISO (1992) ISO 5492:1992 Sensory Analysis Vocabulary. International Standards Organization, Genève (CH).
- ISO (2004) ISO 4120: Sensory analysis Methodology Triangle Test. International Standards Organization, Genève (CH).

Appendix 1 – Sampling, handling and storage of the crop

Sampling of raw materials

A reliable objective random sampling procedure should be used to eliminate subjective effects on the part of the sampler, prevent cross contamination between samples from different unit plots, and reduce to a minimum the effects of variations inherent in growing crops. Although the methods rely basically on random procedures, it may be necessary to use a stratified, rather than a simple, random pattern of sampling, the stratification being on the basis of, for instance, row, compass orientation or aspect (e.g. fruit trees), height of produce on the plant in relation to maturity (e.g. tomatoes which mature from the bottom upwards), prevailing wind or slope of ground. There may also be variations due to, for example, uneven distribution of chemicals both within and over the plant and over the crop as a whole.

The order in which plots are sampled is often important in minimizing the effects of time over the period in which the samples are taken. For instance, a sudden change in light intensity may radically alter the sugar composition of a vegetable such as spinach or tomatoes. Where there are a number of blocks in a trial, the method of sampling should ensure that variation *within* blocks is minimized, by sampling one completed block at a time. In practice, it may be desirable to deal with the control plot(s) within a block first, to eliminate as far as possible any risks of contamination. Trials should not be sampled or harvested treatment by treatment. In general, samples should not be collected when they are wet with dew or rain. Samples taken should be representative of the plot in terms of size, maturity and other physical characteristics.

For the test method recommended in Appendix 2, the requirement of equal amounts of control and treated material for each individual test will give rise to a proportionately large bulk of control material when several treatments, or levels of a treatment, are included in a trial. This should be obtained by taking the required number of control samples in a standard manner rather than by obtaining a large, atypical sample, which has to be handled and stored in different-sized containers from those used for the treated sample(s).

Each crop, cultivar and site may require different sampling procedures, and specialist advice may be needed on the most appropriate procedure to ensure that samples are not atypical of commercial produce.

Hands, containers, tools, machinery, etc. should always be thoroughly cleaned before sampling or handling control material and between taking each sample from the treated plots. For example, treatments applied as a dust may easily be transferred in dry weather from one plot to another. Adequate cleaning facilities should, therefore, be provided. All samples from a trial should be handled in an identical manner and should at all times be shaded from direct sunlight.

Handling of raw materials

Packing

The packing method should give adequate physical protection. If necessary, easily damaged fruits or vegetables such as tomatoes should be individually packed. Containers should be free from contamination, i.e. thoroughly cleaned to remove the risk of chemical, physical and bacteriological contamination, particularly if the test material is to be stored in an unprocessed form. The packing material should not contaminate the samples either physically or chemically. The formation of harmful micro-climates should be avoided, e.g. non-ventilated polythene bags and some types of plastic containers can lead to sweating of the samples. Samples in containers with high thermal insulation properties can reach excessive temperatures. In general, packing in shallow layers is preferable to bulk packing, for both physical protection and regulation of temperature.

Transport

Time in transit should be kept to a minimum. During transport, the samples should be under the personal supervision of a responsible person, and should not be exposed to any risk of external contamination, extremes of heat, etc. It is strongly recommended that public carriers or normal freight-handling facilities should not be used. Transport should be equivalent to the best practice of the food industry and chilled refrigerated transport should be used where possible.

Storage

All raw materials for taint tests should be processed as soon as possible after harvesting. This is particularly important for highly perishable materials such as vined peas, strawberries, etc. Some materials such as potatoes, carrots and apples may have to be stored for varying periods before taint testing or processing. In such cases, the storage conditions should be in accordance with the best commercial practice and should be agreed with the competent authorities. In some cases, it is commercial practice to store raw material in a frozen condition (e.g. -18°C) before manufacture into jam or canned products. Where the practice is a commercially based one, frozen storage is suitable for storing material prior to taint testing.

Processing

Raw material for taint tests should be treated in a manner comparable with recommended commercial practice. For example, strawberries for jam making are generally washed before processing, unlike raspberries and black currants which are processed unwashed. Similarly potatoes and carrots should be peeled in a manner which simulates commercial conditions as closely as possible. It is important that all equipment is thoroughly cleaned between handling different samples. Freezing and heat procedures (canning, jam making and juicing) should be carried out in a standard manner and should reflect commercial process operations. The foods products concerned should conform to any legal standards applicable.

Storage of processed material

Processed materials should be stored under conditions closely similar to those used in commercial practice. Length of time of storage will probably vary since the build up of material for taint tests during the growing season may be more rapid than the completion of the taint tests. The minimum period of storage for taint test purposes is one month for heat-processed products and one week for frozen products. The maximum storage period is the same as the normal commercial shelf-life for the product. This will vary with the crop and processing method.

Appendix 2 – Tasting tests

Method of tasting

The basic method of tasting should be as simple as possible but should also be as accurate as the conditions of the test allow. It is important that proper care be taken to avoid possible sources of bias in carrying out these tests (BSI, 1989; ASTM, 1996), For these reasons, the triangle test is suggested as the standard method for simple taint test work (ISO, 2004). In the triangle test, the assessor is presented with three coded samples, two of which are the same (either control A or treated material B) and one which is different (B or A, respectively). Samples should be presented equally often in each of the six possible orders, ABB, BAB, BBA, AAB, ABA and BAA. The assessor is asked to pick out the odd sample of the three, distinguishing by flavour (including odour) only. At any tasting session two triangle tests may be carried out to increase the rate at which results are obtained.

The triangle test permits a decision only on whether or not the control and treated samples differ. When they do, good methods of determining whether or not a taint has been introduced do not exist, mainly because of the difficulty of defining 'taint' without recourse to hedonic aspects (ISO, 1992) of flavour which demand for their adequate investigation large panels of assessors fairly representative of the consuming public. Trained selected assessors, as recommended here, are more aware of the variations that exist in the natural flavours of crops and food products and are generally better able to express their sensory responses. For these reasons, triangle tests are supplemented by asking the assessors at the time of the triangle test to describe any difference in flavour they may find, and to note the presence of any 'taint'. A treated sample may be 'preferred' to the control, since the treatment may have suppressed a foreign flavour introduced by the pest that it was designed to control. Also, minority reports of unexpected flavours are important, even when the overall result is of no significant difference. Differences between individuals in sensitivity to particular flavours are not uncommon and if such minority reports occur the test should be repeated. If even one assessor consistently finds any aberration this should be recorded and a more intensive investigation carried out.

Although clear cases of taint will be readily distinguished by the descriptions or reactions of individual assessors, there is an intermediate area in which the distinction between 'change of flavour', 'foreign flavour', 'off flavour' and 'taint' is unclear. It is not possible to recommend a procedure which will distinguish clearly between these conditions in marginal cases. However, it is also possible for a subtle change of flavour, not in itself detrimental, to be an early indication of a more serious change that might develop during storage or manufacture of a derived product.

In most cases, an unequivocal result will be obtained. In cases of doubt, and if repeated testing does not clarify the issue, reference should be made to recognized authorities. More fundamental studies of flavour, and especially of its sensory assessment, are needed. It is hoped that growing experience of the effects of agricultural chemicals on the flavour of crops will gradually lead to an improvement of this position, though it is possible that definitions of 'taint' may eventually become specific to each major foodstuff.

Suitability of assessors

Because the possible flavours or taints arising from the use of new chemicals are not known, the selection of a panel on the basis of their sensitivity to a taint is not possible. The panel should, therefore, be composed of persons who, from experience, have shown their ability to discriminate consistently between flavours of the products under test. An assessor whose sense of taste is temporarily impaired, e.g. by a cold, should be excluded.

Number of assessors

The number of persons required for tasting tests, and the number of times they are required to taste each set of samples will vary according to the type of test. The number of assessors required for triangle tests, which are dealing with a wide range of products and flavours, should not be fewer than 10 and should preferably be more. Preferably 18–24 assessors should be used, undertaking tasting once only.

The international standard for the Triangle test (ISO, 2004) recommends that, where testing for similarity of two samples is the objective, at least 30 assessors are needed. This standard proposes that the test objective is to differentiate samples of produce that have been treated or not with a plant protection product. Where testing for difference is the objective, ISO (2004) recommends that at least 18 assessors are needed.

This approach carries the risk that, with a limited number of assessors, some differences may be missed, and some differences that do not exist may be incorrectly perceived. For increased confidence, testing could be conducted for similarity between the treated and untreated. Reference to the ISO standards would be appropriate.

Place of tasting

Taste tests should be conducted in a place from which all outside influence can be excluded. The best conditions are usually those of individual tasting booths where each assessor may examine the samples without distraction. The booths should be of a neutral colour throughout, and contain the bare necessities for the test to take place without interruption. Spoons, recording forms, writing materials and palate cleansers, where required, should be provided.

Standard illumination should be used when necessary. Suitable coloured lighting must be used to mask variations in the colour of the samples.

Time of tasting

The time of day at which tasting takes place will depend on the work of those conducting tests and the number of tests to be done. Some workers believe that more reliable data will result from morning tests than afternoon ones. It is not uncommon, however, to conduct tests in both the morning and afternoon. Tests should be held at such times that the assessors are neither replete nor hungry. If midmorning and mid-afternoon breaks for coffee or tea are permitted, testing should take place before rather than immediately after these times.

Preparation of samples

The preparation of treated samples for tasting should be identical to that for the controls. Samples may be presented in the form in which they are processed or macerated to a puree. If pureed samples are used, the total contents (solid matter plus syrup or brine) of the container should be macerated to a puree but not so thoroughly that fruit seeds are disintegrated. Maceration should be used to blend the sample, small pieces of vegetable being preferable to a smooth over-macerated glutinous paste.

It is recognized that the palate is more sensitive to flavour difference in warmed samples (55°C) than ones at room temperatures. Nevertheless, some authorities feel that samples should be tasted at the temperature at which they are normally consumed. Quick-frozen vegetables should be cooked in a minimum, but standard, amount of water and salt until they are tender and palatable. A proportion of the cooking liquor should be used if they are to be macerated and the samples should be tasted warm. Quick-frozen fruits should be brought to room temperature or warmed by immersion of the containers in hot water, macerated (where applicable) and tasted at room temperature or after warming. Sugar should be added to those samples which were not quick frozen in dry sugar or syrup. Canned fruits and vegetables should be macerated (where applicable) and tasted at room temperature or after warming in a container in hot water. Jams should be tasted either at room temperature or after warming. Maceration is unnecessary but the jam should be stirred or mashed to ensure that the sample is reasonably homogenous. Fruit or vegetable juices should be mixed thoroughly by shaking or stirring. They should be tasted either at room temperature or after warming.