FOOD STANDARDS
Australia New Zealand
Te Mana Kounga Kai – Ahitereiria me Aotearoa

06/03
18 December 2002

FINAL ASSESSMENT REPORT

APPLICATION A404

LACTOPEROXIDASE SYSTEM

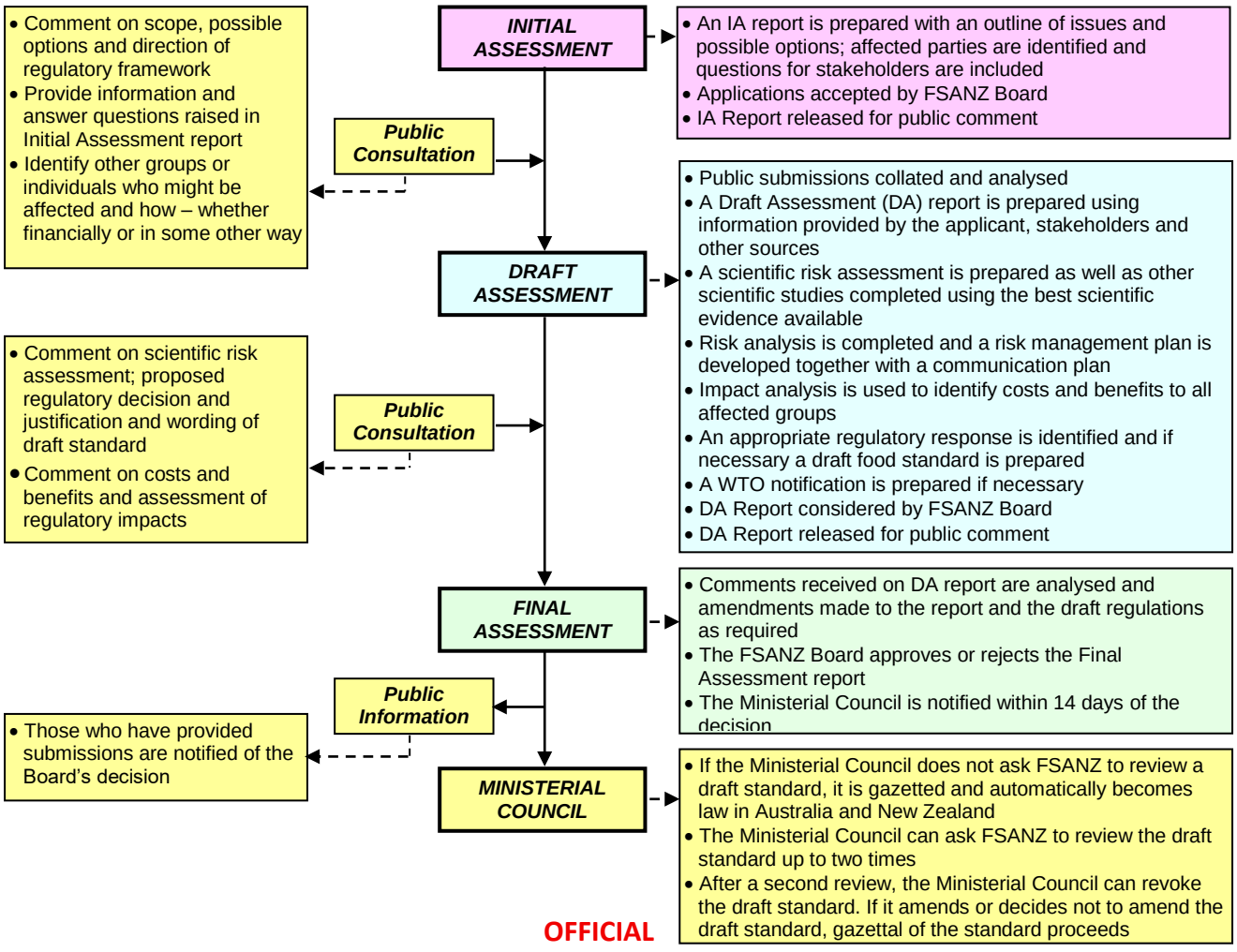
FOOD STANDARDS AUSTRALIA NEW ZEALAND (FSANZ)

FSANZ's role is to protect the health and safety of people in Australia and New Zealand through the maintenance of a safe food supply. FSANZ is a partnership between ten governments: the Commonwealth; Australian States and Territories; and New Zealand. It is a statutory authority under Commonwealth law and is an independent, expert body.

FSANZ is responsible for developing, varying and reviewing standards and for developing codes of conduct with industry for food available in Australia and New Zealand covering labelling, composition and contaminants. In Australia, FSANZ also develops food standards for food safety, maximum residue limits, primary production and processing and a range of other functions including the coordination of national food surveillance and recall systems, conducting research and assessing policies about imported food.

The FSANZ Board approves new standards or variations to food standards in accordance with policy guidelines set by the Australia New Zealand Food Regulation Ministerial Council (Ministerial Council) made up of Commonwealth, State and Territory and New Zealand Health Ministers as lead Ministers, with representation from other portfolios. Approved standards are then notified to the Ministerial Council. The Ministerial Council may then request that FSANZ review a proposed or existing standard. If the Ministerial Council does not request that FSANZ review the draft standard, or amends a draft standard, the standard is adopted by reference under the food laws of the Commonwealth, States, Territories and New Zealand. The Ministerial Council can, independently of a notification from FSANZ, request that FSANZ review a standard.

The process for amending the *Food Standards Code* is prescribed in the *Food Standards Australia New Zealand Act* 1991 (FSANZ Act). The diagram below represents the different stages in the process including when periods of public consultation occur. This process varies for matters that are urgent or minor in significance or complexity.



Final Assessment Stage

The Authority has now completed two stages of the assessment process and held two rounds of public consultation as part of its assessment of this application. This Final Assessment Report and its recommendations have been approved by the FSANZ Board and are now being reviewed by the Australia and New Zealand Food Regulation Ministerial Council (ANZFRMC).

If accepted by ANZFRMC, a change to the *Food Standards Code* is published in the *Commonwealth Gazette* and the *New Zealand Gazette* and adopted by reference and without amendment under Australian State and Territory food law.

In New Zealand the New Zealand Minister for Health gazettes the food standard under the New Zealand Food Act. Following gazettal, the standard takes effect 28 days later.

Further Information

Submissions

No submissions on this matter are sought as the Authority has completed its assessment and the matter is now with the Australia and New Zealand Food Regulation Ministerial Council for consideration.

Further information on this application and the assessment process should be addressed to the Standards Liaison Officer at Food Standards Australia New Zealand at one of the following addresses:

Food Standards Australia New Zealand
PO Box 7186
Canberra BC ACT 2610
AUSTRALIA
Tel (02) 6271 2222
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Food Standards Australia New Zealand
PO Box 10559
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Assessment reports are available for viewing and downloading from the FSANZ website www.foodstandards.gov.au . Alternatively paper copies of reports can be requested from the Authority's Information Officer at either of the above addresses or by emailing info@foodstandards.gov.au including other general enquiries and requests for information.

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Executive Summary

The application (A404) from Tatua Cooperative Dairy Co Ltd was to permit the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids on meat. Lactoperoxidase and sodium (and potassium) (iso)thiocyanate are components of a lactoperoxidase system (LPS) with the function of inhibiting bacteria. The applicant advised in a letter dated 18 December 2000 that only sodium thiocyanate was used. The other components of LPS - glucose oxidase and glucose are permitted already in the joint Australia New Zealand Food Standards Code as processing aids.

The Australia New Zealand Food Authority (ANZFA) to Food Standards Australia New Zealand (FSANZ) transitional requirements for an application at full (draft) assessment stage have been followed. The Authority has not received additional submissions in relation to this application and to date it has not been notified of any Ministerial Council policy guidelines relevant to this application.

Permitting the use of these processing aids to levels determined by Good Manufacturing Practice may be of public health benefit to consumers and reduce the incidence of food-borne illness when used as an additional hurdle in a food safety system for the treatment of meat. At the levels of use proposed in the application neither the components of the lactoperoxidase system, nor the intermediary products, pose a significant risk to human health, apart from the potential for adverse reactions to milk proteins. Lactoperoxidase is a milk derived protein and the LPS system also contains some other milk proteins. Consumers allergic to milk protein will need to be made aware of its presence on meat products. Any risk to such consumers, given that meat products may not normally be considered as carrying any risk of exposure to milk allergy proteins, will be adequately addressed by the labelling requirement.

It is recommended that consumers be informed by appropriate labelling of meat and meat products for the presence of milk proteins as required by Standard 1.2.3.

Since draft assessment FSANZ has decided:

(a) because there is a mandatory labeling requirement in Standard 1.2.3 at clause 4, that this requirement should not be repeated in the meat product standard; and

(b) for public health and safety reasons, an editorial note cross referencing to the labeling standard should be included in clause 14 of Standard 1.3.3 for processing aids.

Statement of Reasons

FSANZ has agreed to adopt the draft variation proposed in A404 for the following reasons:

- Permitting the use of these processing aids to levels determined by Good Manufacturing Practice may be of public health benefit to consumers and reduce the incidence of food-borne illness when used as an additional hurdle in a food safety system for the treatment of meat.

- At the level of use proposed in the application neither the components of the lactoperoxidase system, nor the intermediary products pose a significant risk to human

health for the majority of the population.

- Consumers allergic to milk protein will need to be made aware of its presence on meat products. While meat products are not normally considered to carry any risk of exposure to milk proteins, any potential risk to consumers will be adequately addressed by the labelling requirement.

- The proposed changes to Volume 2 of the *Food Standards Code* are consistent with the section 10 objectives of the *Food Standards Australia New Zealand Act 1991*.

- The Regulatory Impact Statement indicates that, for the preferred option, namely, to approve the use of lactoperoxidase from bovine milk and sodium thiocyanate, the benefits of the proposed amendment outweigh the costs.

1. Introduction

The Application (A404) from Tatua Cooperative Dairy Co Ltd was to permit the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids on meat. The applicant confirmed that only sodium thiocyanate is to be used. Lactoperoxidase and sodium (and potassium) (iso)thiocyanate are components of a lactoperoxidase system (LPS) with the function of inhibiting bacteria. The other components of LPS - glucose oxidase and glucose are permitted already in the joint ANZFSC as processing aids.

1.1 Transitional Requirements

This Application reached Full (Draft) Assessment stage under the operation of the *Australia New Zealand Food Authority Act 1991* (ANZFA Act), and will be finalised in accordance with the provisions of the *Food Standards Australia New Zealand Act 1991* (FSANZ Act).

FSANZ has therefore been required to:

1. give the Applicant the opportunity to (by 29 July 2002) request deferral of consideration of the application in order to provide any additional information;

2. give notice under section 14 of the FSANZ Act; and

3. review the Full (Draft) Assessment having regard to any new submissions received in response to the above notice as well as any written policy guidelines that have been notified by the Ministerial Council.

2. Regulatory Problem

Standard 1.3.3 - Processing aids regulates the use of processing aids in food manufacture, prohibiting their use unless there is a specific permission within the Standard. There is currently no permission for lactoperoxidase from bovine milk or for sodium thiocyanate.

Standard 1.2.3 – Mandatory Warning and Advisory Statements and Declarations requires in clause 4 the mandatory declaration of certain substances, including milk products, in food. The presence of milk products must be declared when present as an ingredient, compound ingredient, food additive or processing aid or components of these.

3. Objectives

The objective of this Application is to determine whether it is appropriate to change the *Food Standards Code* to allow the use of the lactoperoxidase system.

In developing or varying a food standard, FSANZ is required by its legislation to meet three primary objectives which are set out in section 10 of the FSANZ Act. These are:

- the protection of public health and safety;
- the provision of adequate information relating to food to enable consumers to make informed choices; and
- the prevention of misleading or deceptive conduct.

In developing and varying standards, FSANZ must also have regard to:

- the need for standards to be based on risk analysis using the best available scientific evidence;
- the promotion of consistency between domestic and international food standards;
- the desirability of an efficient and internationally competitive food industry;
- the promotion of fair trading in food; and
- any written policy guidelines formulated by the Ministerial Council.

4. Background

4.1 Historical Background

On the 30 November 1999, an application (A404) was received from Tatua Cooperative Dairy Co Ltd to permit the use of the lactoperoxidase system (LPS) as a processing aid within Standard A16 (Standard 1.3.3 in the Code for the treatment of:

- meat and meat products (including poultry),
- fish and fish products and
- milk and milk products.

The Applicant claimed that LPS treatment inhibits some pathogenic and spoilage bacteria in the food. The components of LPS are the enzymes lactoperoxidase and glucose oxidase, plus glucose and a source of (iso)thiocyanate ions, either potassium or sodium thiocyanate.

The enzyme glucose oxidase and glucose as a food already have permissions in the *Australia New Zealand Food Standards Code* as generally permitted processing aids. Hence the application is for the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids in meat.

Between 23 February 2000 and 5 April 2000, the Preliminary Assessment Report (Initial Assessment) for A404 was released for public comment. Five submissions were received in response to the public consultation. On 27 June 2000, requests for further information were sent to the Applicant.

The clock remained stopped on the application until 25 January 2002 when a response from the Applicant was received by the then ANZFA.

ANZFA prepared a Full/Draft Assessment Report for consideration and submissions for public comment closed 7 August 2002. Nineteen submissions were received, a number of these being reviews from Food Science graduates at the University of Auckland.

No additional submissions were received in response to the section 14 notice required under the transitional provisions of the Act.

5. Relevant Issues

5.1 Foods in which lactoperoxidase components would be used

Initially the Applicant sought permission for the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids to be applied to a broad range of foods including meat, fish, milk and their products. The published information on the effectiveness of these processing aids on a wide range of foods is limited. The Applicant therefore limited the scope of the application to meat and meat products.

5.2 Level of usage of the processing aids

In order for the LPS system to be effective as an antimicrobial processing aid the components must be applied at appropriate levels. These are:

- Lactoperoxidase, 800 to 2800 U per kg meat;
- Sodium (or potassium) thiocyanate, 30 to 40 mg per kg meat as the thiocyanate ion, SCN^-;
- Glucose oxidase, 150 to 300 U per kg meat; and
- Glucose 120 to 160 mg per kg meat;

The individual components of the system are effective when used together at these levels. Thus application at the above levels will be commensurate with Good Manufacturing Practice (GMP).

5.3 Processing aid function as an antibacterial agent

As explained in the Food Technology Report (Attachment 3) the applicant is seeking approval of LPS as a processing aid that could contribute to a hurdle system that will minimise the risk to consumers of pathogens on meat. Hurdles are factors, conditions or processing steps that limit, retard or prevent microbial growth and/or reduce the microbial load but which cannot by themselves keep microbiological hazards under control. This definition can be applied to LPS. It is important to note that LPS will reduce but not eliminate pathogens present on the meat surface and that these pathogens will not always be present as the meat industry is actively engaged in a number of strategies to minimise carcase contamination.

The ions generated by activation of LPS damage bacterial membranes and impair metabolic enzymes. As there is some variation in the structure of the bacterial membranes associated with the cell walls of the various bacterial species, LPS will show variable effects related to which bacterial species are present in the food being treated. As one of the antibacterial effects is to impair metabolic enzymes, these effects may be manifest only when bacteria are growing. If the bacteria are not actively metabolising at the time of treatment with LPS the antibacterial effects will be lessened. Activity against cold tolerant bacteria such as *Listeria monocytogenes* is thus more pronounced than other bacterial species, when LPS is applied to chilled meat.

Treatment with LPS may inhibit bacteria present on food. The extent of the inhibitory effects will be related to the bacterial species present and the temperature of the food.

5.4 Toxicity of lactoperoxidase system components

The toxicological assessment (Attachment 4) concludes that at the levels of use proposed in the application neither the components of the lactoperoxidase system, nor the intermediary products, pose a significant risk to human health, provided consumers allergic to milk protein are aware of its presence.

There are several features that support the safe use:

- the occurrence of all three components in human and animal systems;
- the high levels of thiocyanate present naturally in human saliva and gastric juice;
- the presence of the intermediate compound OSCN- in human saliva; and
- the short-lived nature of intermediate compounds.

5.5 Residual milk protein and labelling for potential allergenicity

The lactoperoxidase enzyme is a milk-derived protein and the enzyme preparation contains some other milk proteins. The amount present will depend on the level of purity of the lactoperoxidase. A high-grade commercial lactoperoxidase will contain 10% of other milk protein (OMP). At the levels of proposed use (2800 U of lactoperoxidase per kg of meat) the maximum amount of potentially allergenic milk protein that could be present would be about 1 mg per kg of meat.

Professor Steve Taylor from the Food Allergy and Research and Resource Program at the University of Nebraska was commissioned by the applicant to provide an expert opinion on the potential allergenicity of lactoperoxidase. His conclusion (report dated January 21, 2002) is that "lactoperoxidase is not a known allergen and the presence of known allergens in commercial lactoperoxidase seems insufficient to elicit allergic reaction in the vast majority of milk-allergic individuals. Weak evidence exists to suggest that lactoperoxidase may be capable of sensitising susceptible individuals. However given the low levels of predicted dietary exposure to lactoperoxidase, that possibility seems unlikely." In addition his assessment of the 10% OMP present is that the exposure would be below "the lowest observed adverse effect level for milk protein encountered in clinical challenge tests of highly sensitive individuals of 0.6 mg".

While the level of milk protein may be low, there would nevertheless be the need to meet the requirements of Standard 1.2.3 with regards to labelling. Standard 1.2.3 requires that where foods contain milk and milk products these must be declared on the label, displayed in connection with the food or provided to the purchaser on request.

5.6 Use of lactoperoxidase system components in other countries

Codex Standard (CAC/GL 13-1991) provides for the use of the lactoperoxidase system for the stabilisation of milk, although refrigeration remains the method of choice for safe milk transport. When applied to dairy products, the major component of the system,

lactoperoxidase, is present in the milk. The system is activated by the addition of thiocyanate and hydrogen peroxide in the form of sodium percarbonate.

The activation of the LPS system in raw milk is used to prevent undue bacterial multiplication during collection and transport to the dairy processing plant in countries where refrigeration may not be feasible due to technical or economic reasons.

In France, the Ministry of the Economy of Finance and Industry gave a one-year permit for the addition of LPS to the brine "destined for the production of smoked salmon" in April 1998. No information has been made public from that work of which FSANZ is aware.

5.7 Public health benefits of LPS treatment

There is no published evidence of the effectiveness of LPS in reducing or eliminating pathogenic or spoilage bacteria from the surface of meat other than laboratory studies on ground meat. If pathogenic bacteria are present on the meat they could cause food-borne illness if the meat or a meat product made from that meat is inadequately cooked. Contaminated meat may also act as a source of pathogens that could cross-contaminate other foods which could then become hazardous.

Studies commissioned by the Applicant and undertaken by the Meat Industry Research Institute and provided to the then ANZFA, demonstrated a variable effect of LPS with the greatest reductions for the cold tolerant bacteria (*Listeria monocytogenes* and spoilage bacteria) and minimal effects against *E. coli O157:H7*. In these studies it was necessary to add the microbial contaminants at high levels in order to have a measurable effect. Natural contamination occurs very infrequently under modern day meat processing requirements.

Lactoperoxidase based systems (LPS) have been researched and investigated for application to dairy products and milk as the lactoperoxidase naturally occurs in milk. There is very limited information in the literature of non-dairy applications and thus the applicant was unable to supply published information that demonstrated effectiveness under the proposed conditions of use.

However the outcome of research subsequently undertaken by the applicant has been provided to FSANZ but this research has not yet been published by the applicant. Research on the LPS system has also been undertaken and published by researchers from Otago University since this application was received. This research provides some information on the activity of LPS using a minced meat laboratory model system and in broth cultures.

6. Regulatory Options

6.1 Option 1: Maintain the *status quo* and not permit the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids on meat.

This option would not be consistent with the section 10 objectives. Neither lactoperoxidase and sodium (and potassium) thiocyanate at the levels of use proposed in the application, nor the intermediary products, pose a significant risk to human health, provided consumers allergic to milk protein are aware of its presence.

Not giving permission for the processing aids for use on meat and meat products would be contrary to the objective of allowing food standards that protect public health and safety, because the LPS system provides an additional hurdle for the growth of some pathogens.

6.2 Option 2: Amend Standard 1.3.3 to permit the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids on meat.

This option is consistent with the section 10 objectives as it permits meat to be treated with lactoperoxidase and sodium (and potassium) thiocyanate as processing aids as part of LPS in order to reduce the numbers of, or inhibit the growth of bacteria on the surface of the meat. . There is no specification for potassium thiocyanate in the publications listed as sources in Standard 1.3.4. The applicant confirmed that only sodium thiocyanate is to be used. It is therefore not necessary to approve potassium thiocyanate in Standard 1.3.3.

There are public health benefits in that the treatment introduces an additional hurdle and meat would be less likely to be a source of food-borne illness in circumstances where the meat was inadequately heat-treated.

7. Impact analysis

7.1 Industry

Amendment of the Code will allow suppliers of lactoperoxidase and sodium thiocyanate to market their product as processing aids to the meat industry.

Amendment of the Code will permit the use of lactoperoxidase and sodium thiocyanate as processing aids for use in the LPS system for meat and meat products. The use of LPS will provide the meat industry with an additional treatment that can be used as a hurdle to reduce pathogen contamination on meat and meat products. The reduction in bacterial levels may be proportional to the level of contamination present and may not be equal for all bacterial species. Some significant meat borne pathogens are unlikely to be reduced significantly by treatment, limiting the value of the treatment to the meat industry. The meat industry will be permitted the opportunity to use an innovative and safe treatment.

Meat processors using LPS will need to set up systems for applying LPS to meat and meat products correctly and for identifying treated meat and meat products and for keeping it separate from untreated meat. They will need to be able to identify this meat in the distribution system so that the sellers of the treated meat, including the food service industry will know that the meat and meat products must be labelled or the consumer made aware that the meat has been treated.

7.2 Consumers

Consumers will have access to safer meat products resulting from the use of an additional hurdle treatment in accordance with Good Manufacturing Practice. The LPS treatment has the potential to reduce the incidence of illness from contaminated meat and meat products, although this is not a major source of food-borne illness in Australia or New Zealand. Treatment with LPS will not however lessen the need for good food handling practices in the home or food service industry.

LPS treated meat and meat products will be required to be appropriately labelled for allergenicity. The level of milk protein that would be present in the meat would be low and expert opinion has indicated that it is unlikely that the levels would cause serious reactions if accidentally ingested by a sensitive person. The levels expected in most products are comparable with those of hypoallergenic infant formula.

Treated meat and meat products could potentially cost more for consumers as processors seek to recoup the costs of undertaking the LPS treatment.

7.3 Overall analysis

Permitting the use of lactoperoxidase and sodium thiocyanate as processing aids as part of the LPS by industry will provide an additional hurdle for bacterial pathogens that meat processors may include in a food safety system.

This may have an impact on the incidence of food-borne illness in consumers but the reduction that would be achieved cannot be readily measured and would not be expected to be great. The need to have equipment to apply LPS to the meat and to track treated meat through further processing and the distribution system may limit the use of LPS by industry.

While the presence of milk and milk proteins in some processed meat products may be anticipated by consumers allergic to these substances, the presence of these allergens in other meats such as mince or blocks of meat may not be expected by consumers. Hence the treated meat will be required to be appropriately labelled to inform consumers. The additional costs of labelling may have an impact on the cost of production of the meat and the sale price.

8. Consultation

8.1 Initial Assessment

At the Initial Assessment, public comment was sought on the use of lactoperoxidase and sodium (and potassium) thiocyanate as processing aids for a wide variety of foods.

Submissions raised concerns about the use of a milk product in foods that would not be expected to contain milk proteins. The scope of the application has since been limited to the use of the processing aids for the treatment of meat only that would be appropriately labelled to disclose the presence of milk protein.

Submissions also queried the need for these processing aids for the purpose of antibacterial treatment of meat in Australia and New Zealand, which have adequate refrigerated storage for food. The applicant does not request permission for the use of LPS as an alternative to good manufacturing practice or good hygienic practice but as an additional hurdle as part of a food safety system that may also include refrigeration. The food technology report (Attachment 3) addressed this matter.

Submissions also requested an assessment of the possibility of toxic residues on treated food. The toxicological assessment (Attachment 4) addressed this matter.

8.2 Draft Assessment

Of the 19 submissions received, 13 were essentially reviews of the Draft Assessment and were undertaken by students from Auckland University. In most of the student submissions no view was expressed as to a preferred option. Where a review was expressed or implied, Option 2, to allow the amendment was favoured. A number of the student submissions recommended further research on efficacy or did not believe that sufficient evidence of efficacy had been provided. However the applicant has demonstrated that LPS does have antimicrobial activity and while further research would provide information on additional uses and efficacy of LPS, it would not be pertinent to the current application to amend Standard 1.3.3. Issues around consumer education and reaction to the use of a milk derived processing aid on meat were raised in one submission. However, labelling of treated meat should provide sufficient information to people making purchasing decisions, given that there are other milk derived products that may be found in meat products.

Of the remaining six submissions, five favoured the amendment, although one from the AFGC submitted that the use of LPS should be extended for food generally and not restricted to meat, in accordance with the principles developed for the review of food regulations. However the wider usage has not been assessed and until there is relevant data available to confirm a technological need for its use in other food matrices it would not be appropriate to allow wider use.

The one dissenting submission was from Food Technology Association of Victoria which repeated the submission made at Initial Assessment and did not support the amendment to allow the use of LPS on meat. All the issues raised had however been fully addressed in the Draft Assessment, which the submitter had not read in detail when spoken to.

The submissions in general support the proposed amendment and no scientific evidence has been provided to the contrary by the submissions.

No submissions were received in response to the section 14 notice required under the transitional provisions of the Act.

8.3 World Trade Organization Notification

Australia and New Zealand are members of the WTO and are bound as parties to WTO agreements. In Australia, an agreement developed by the Council of Australian Governments (COAG) requires States and Territories to be bound as parties to those WTO agreements to which the Commonwealth is a signatory. Under the agreement between the Governments of Australia and New Zealand on Uniform Food Standards, FSANZ is required to ensure that food standards are consistent with the obligations of both countries as members of the WTO.

In certain circumstances Australia and New Zealand have an obligation to notify the WTO of changes to food standards to enable other member countries of the WTO to make comment. Notification is required in the case of any new or changed standards which may have a significant trade effect and which depart from the relevant international standard (or where no international standard exists).

This matter does not need to be advised to the WTO as a TBT or a SPS Notification because the proposed change to the regulation is optional for manufacturers and unlikely to significantly effect trade.

9. Conclusions

It is concluded that the *Food Standards Code* be amended to include lactoperoxidase and sodium thiocyanate as processing aids in Standards 1.3.3 for the following reasons:

- Permitting the use of these processing aids to levels determined by Good Manufacturing Practice may be of public health benefit to consumers and reduce the incidence of food-borne illness when used as an additional hurdle in a food safety system for the treatment of meat.

- At the level of use proposed in the application neither the components of the lactoperoxidase system, nor the intermediary products pose a significant risk to human health for the majority of the population.

- Consumers allergic to milk protein will need to be made aware of its presence on meat products. While meat products are not normally considered to carry any risk of exposure to milk proteins, any potential risk to consumers will be adequately addressed by the labelling requirement.

- The proposed changes to Volume 2 of the *Food Standards Code* are consistent with the section 10 objectives of the *Food Standards Australia New Zealand Act 1991*.

- The Regulatory Impact Statement indicates that, for the preferred option, namely, to approve the use of lactoperoxidase from bovine milk and sodium thiocyanate, the benefits of the proposed amendment outweigh the costs.

ATTACHMENTS

1. Draft variation to *Food Standards Code*
2. Summary of Public submissions received at Initial and at Draft Assessment
3. Food technology report
4. Toxicology report

Attachment 1

DRAFT VARIATION TO THE *FOOD STANDARDS CODE*

APPLICATION A404 – LACTOPEROXIDASE SYSTEM

To commence: on gazettal

[1] ***Standard 1.3.3*** *of Volume 2 of the Food Standards Code is varied by –*

[1.1] *inserting immediately before the* Table to clause 14 -

Editorial note: Where meat has been treated using lactoperoxidase the mandatory labelling requirements in clause 4 of Standard 1.2.3 apply.

[1.2] *inserting in the* Table to clause 14 *in alphabetical order* -

Lactoperoxidase from bovine milk EC [1.11.1.7]	Reduce and/or inhibit bacterial population on meat surfaces	GMP

[1.3] *inserting in the* Table to clause 14 *in alphabetical order* -

Sodium thiocyanate	Reduce and/or inhibit bacterial population on meat surfaces	GMP

Attachment 2

SUMMARY OF PUBLIC SUBMISSIONS RECEIVED

A404 – LACTOPEROXIDASE SYSTEM

INITIAL ASSESSMENT

Food Technology Assoc of Victoria	• No technological justification • Refrigeration and pasteurisation adequate in Australia • Actually a preservative effect • Actually a hydrogen peroxide source • Would not be detectable as a processing aid • Queries if consideration has been given to residual thiocyanate
Barbara Baragwanath, Orakei	• Possible serious dangers posed to health by additives in processed foods
Natalie Baragwanath, Auckland	• Consumers could be concerned that a cow's milk enzyme is being added to meat and meat products
Ministry of Health	• Insufficient evidence that this is a processing aid rather than additive function. • Insufficient information on proposed use • Proposed use is not the same as that permitted by Codex • Assessment should not contain reference to use of the lactoperoxidase system as an alternative to pasteurisation as this is not correct • Will need to review the toxicological data when the application is at full assessment
InforMed Systems Ltd (John Birkbeck)	• Supports application • No adverse implications for health and safety • Useful in food processing

DRAFT ASSESSMENT

Food Technology Assoc of Victoria (David Gill)	• Prefer option 1 – maintain the status quo because: • Poor effectiveness compared with other agents • An allergy declaration should be required • No technological justification • Refrigeration and pasteurisation adequate in Australia • Actually a preservative effect • Actually a hydrogen peroxide source • Would not be detectable as a processing source through labelling • Queries if consideration has been given to residual thiocyanate • Uses of milk in a meat product would be unacceptable for some consumers

Australian Food and Grocery Council	• Object to the restriction of use to meat only • Outcome of the restriction is inconsistent with principles established in the Review • FSANZ unnecessarily concentrates on technical efficacy • Supports Option 2 but delete the reference to meat
Shane Hopgood, Food Safety Consultant/Auditor	• Supports amendment
Informed Systems Ltd (John Birkbeck)	• Supports amendment
Fonterra (Joan Wright)	• No objection to Option 2
Doug Watson	• Supports amendment
Hemant Kukde	• Reviews application • No preferred option
Tin Sang Tsang	• Reviews application • No preferred option
Anand R Chordia	• Reviews application • No preferred option
Seema Kumar	• Reviews application • No preferred option
Sharyn Gee	• Concerned as to who would be responsible for consumer education • Preferably not used on domestic meat until consumer reaction established
Jane Agnes Olivares	• Efficacy not fully established • Further research needed before approved
Kuek Tze Lee	• Supports Option 2
Leena Gangal	• Supports Option 2 but believes more research on effectiveness needed before permitted
Santhameena Sivaplan	• Supports Option 2 but believes more research on effectiveness needed before permitted
Mrinalini (Leena) Kukde	• Reviews application • No preferred option
Anusooya Evangaline Ravi	• Reviews application • No preferred option
Surianto Mangiri	• Option 2 preferred
Western Australia Food Advisory Committee (Virginia McLaughlin)	• Supports Option 2

Attachment 3

FOOD TECHNOLOGY REPORT

APPLICATION A404 – LACTOPEROXIDASE SYSTEM

Introduction

The application is for an antibacterial system for addition to meat, other than poultry to provide a food safety hurdle for pathogens. The applicant contends that improperly cooked and stored meat are potent sources of a wide range of pathogens. Expert groups hold this view. For example - "such products have been implicated in a number of recent serious food poisoning outbreaks internationally. In spite of increasingly sophisticated hygiene measures, carcasses, meat cuts, processed meat and fish are still found to be contaminated with pathogens such as *Salmonella spp., E. coli, Campylobacter spp, Yersinia* spp and *Listeria monocytogenes"*(ICMSF 2000).

A report prepared for the Applicant by Dr Andrew Hudson from the Institute for Environmental Science Research identified the level of pathogens associated with red meat in New Zealand and Australia and evaluated the contribution this could make to food-borne illness. He concludes that while not a major source of food-borne illness, red meat can be linked epidemiologically with a number of disease reports. He also notes the very strong link that exists between major outbreaks of food-borne illness in both the U.K. and the USA.

A number of countries have introduced pathogen reduction and critical control point systems for the meat industry. The Food Safety and Inspection Service of the United States Department of Agriculture for example considers this approach reduces the risk for food-borne illness (FSIS, 1998).

LPS in the dairy industry

The lactoperoxidase system (LPS) has been extensively investigated for dairy industry application (Farrag & Marth, 1992). When applied to dairy products, the major component of the system lactoperoxidase is present in the milk. The system is activated by the addition of thiocyanate and hydrogen peroxide in the form of sodium percarbonate. The activation of the LPS system in raw milk is used to prevent undue bacterial multiplication during collection and transport to the dairy processing plant in countries where refrigeration may not be feasible due for technical or economic reasons. The use of lactoperoxidase for the stabilisation of milk has been approved by Codex CAC/GL 13-1991), although refrigeration remains the method of choice for safe milk transport.

Use of the LPS system has been extended to other dairy products at least in the laboratory and there is evidence of significant gains in terms of food safety and keeping quality for cottage cheese and raw milk cheeses (Earnshaw et al, 1989). Dairy products frequently rely on a number of hurdles to ensure safety and shelf life and activation of the naturally occurring lactoperoxidase is one such hurdle.

Non-dairy application of LPS

Extension of LPS to non-dairy foods requires the lactoperoxidase to be added, as it will not be naturally present in the food. There is very limited information in the literature of non-dairy applications. In France, the Ministry of the Economy of Finance and Industry gave a one-year permit for the addition of LPS to the brine "destined for the production of smoked salmon" in April 1998. No information has been made public from that work that FSANZ is aware of.

Some research indicates that LPS may have considerable advantages when used in conjunction with other antimicrobial treatments, as synergistic effects are commonly demonstrable.

Research to demonstrate the effectiveness of LPS in a meat matrix has been undertaken in New Zealand at the University of Otago and at MIRINZ using the Tatua LPS. Not unexpectedly it was observed that the enzymes are most active, and therefore antimicrobial effects are most significant, under temperature conditions that would be associated with product abuse rather than those associated with good hygienic practices in meat processing (Kennedy et al, 2000). While it is possible that meat may at some time be exposed to sub-optimal storage conditions (say >8 degrees C) before consumption, the benefit of the meat having been treated with LPS would be of limited value since the LPS is active only for a short period after application to the meat.

Evidence of effectiveness

The Applicant, at the request of the then ANZFA, undertook trials in which the LPS was applied to meat in a model system which replicated the intended commercial use of the LPS. The applicant has supplied data from research at Otago University (some of which has since been published, Kennedy et al, 2000) as well as trials performed by an independent scientific organisation (Meat Industry Research Institute of NZ) on meat cuts processed and stored in vacuum packs to model normal commercial practices.

To assess the pathogen reduction ability of the system, meat samples were inoculated with *Listeria monocytogenes* or *Escherichia coli* O157:H7. The trials demonstrated decreases in the levels of and/or growth during storage of the inoculated pathogens and of the naturally occurring bacterial populations. As the antimicrobial activity occurs during bacterial growth, the effects of the treatment were most pronounced against bacterial species able to grow at the temperatures used during, and subsequent to LPS treatment. Thus the effects were greater against *L. monocytogenes* and natural spoilage bacteria which are all able to grow at low temperatures, than against *E. coli O157:H7* which survives but does not grow readily under these conditions. Therefore the reduction in growth of *E. coli O157:H7* was minimal. However the treated cells could possible be damaged in such a way as to less likely to cause illness if ingested than bacteria from untreated meat. However as bacteria are able often to repair damage, this may not be a realistic scenario. No evidence was provided on this.

The variation in the naturally occurring populations and population interactions will mean that effects will not be the same in each trial. It must also be taken into account that the artificial inoculations result in levels of contamination with pathogens in excess of what would be expected to occur using good hygienic practices but which are necessary in the trials in order to demonstrate a measurable outcome from the treatment.

Of particular relevance to this application is that the effect of the LPS treatment is a reduction on microbial loads only. This reduction, depending on the bacteria may be as little as 0.5 log for one major meat borne pathogen. This would provide some reduction in potential hazard levels but depending on the initial carcass load, would not reduce the level of care that would need to be taken during subsequent handling, processing and cooking to prevent cross contamination from the meat occurring or to obviate the need for adequate cooking or precautions in the preparation of raw meat dishes.

In conclusion, LPS has been shown to be effective at reducing and inhibiting microbial populations on meat. The effect varies according to the bacterial species present.

Meat as a source of food poisoning organisms

Meat has the potential to carry food poisoning organisms to consumers. The bacteria which constitute a hazard in at least some meat products are *Salmonella sp;* enterohaemorrhagic *Escherichia coli* (EHEC*),* some serovars of *Yersinia enterocolitica, Campylobacter jejuni, Staphylococcus aureus, Listeria monocytogenes, Clostridium perfringens, Clostridium botulinum* and *Bacillus cereus.* Raw meat is also subject to spoilage by a range of microorganisms and is a highly perishable commodity (ICMSF, 2000).

Under current conditions of meat production and processing only small numbers of Salmonella are normally found on carcases but inadequate chilling, storage or transport at temperatures above 8 degrees can permit growth. Outbreaks of salmonellosis can follow from inadequate cooking, mishandling and recontamination. Raw meats can act as a source of cross-contamination of cooked meats, or other foods, in the kitchen or in meat processing plants. Carcasses are considered a relatively minor cause of human Campylobacter infection and are not a source of staphylococcal food poisoning or botulism. Raw meat can be one of the sources of contamination of ready-to-eat processed meats with *Listeria monocytogenes.* Inadequately cooked ground beef contaminated with *E. coli* O157:H7 has caused a number of outbreaks (ICMSF, 2000). Thus of these the pathogens those of most concern are EHECs such as *E. coli* O157:H7 and *Salmonella sp.*

Commercial use of LPS

The applicant is seeking approval of LPS as a contribution to a hurdle system that will minimise the risk to consumers of pathogens on meat. Hurdles are factors, conditions or a processing step that limit, retard or prevent microbial growth and/or reduce the microbial load but which cannot by themselves keep microbiological hazards under control. This definition can be applied to LPS.

It is important to note that LPS will reduce but not eliminate pathogens present on the meat surface. These pathogens will not always be present as the meat industry is actively engaged in a number of strategies to minimise carcase contamination.

Assessment of the benefits of using LPS

Meat is not a direct cause of food-borne illness unless the meat is inadequately cooked or if eaten raw, not prepared appropriately. As LPS treatment only reduces and does not eliminate bacteria, contaminated, treated meat poorly cooked or incorrectly prepared would be potentially less risky to consumers.

However, it is important to note that undercooking of meat, in particular hamburgers has not been a significant source of illness in Australia and New Zealand, unlike some other countries, in particular the USA.

Raw meat may be a source of cross-contamination in both processed meat plants and in catering and home kitchens. The less contaminated raw meat the less likely is cross-contamination. However cross-contamination should not occur in the kitchen if good hygienic practices are in place. Reducing the carriage of *Listeria monocytogenes* transported into small good processing plants would reduce the risk of subsequent contamination of cooked ready-to-eat meats. This could be a useful effect.

Assessment of the risks from using LPS

Meat treated with LPS would remain a potential hazard for food-borne illness if incorrectly cooked and prepared for consumption**.** Treated meat could be considered to be safer than untreated meat but still not sufficiently low risk as to be handled any differently from untreated meat in the processing plant and the food service or home kitchen.

Conclusion

There is limited information relating to the use of LPS as a microbial hurdle for other than dairy products. The available information relating to meat has been generated in association with the current application. At this point in time it is still difficult to define expected outcome of treatment with LPS in terms of which populations will be reduced and by how much, other than an expectation that some effects will occur and that these can be defined as reducing the microbiological population and/or retarding the growth of the microbial populations on meat surfaces.

There are data that demonstrate that carcases and boned meat in Australia and New Zealand may be contaminated with bacterial pathogens. The level of contamination for pathogens such as Salmonella, Campylobacter and *E. coli* O157:H7 is usually <1.5% (Philips et al, 2001a, 2001b).

While processed red meat has been shown to be on occasion (five documented outbreaks in five years) to be the source of food-borne illness in Australia and New Zealand, it is not always clear what the sources of the contamination in each case were and/or what the other factors that contributed to the outbreaks were. In some cases there is evidence of significant mishandling of the meat occurring (Hudson, 2001)

Since the effects of LPS are only relative and not absolute, even LPS-treated meat could still potentially cause food-borne illness if the critical control point failures were of a large magnitude. Thus the potential benefits from LPS are undefined in terms of public health outcomes but not expected to be large in the New Zealand and Australian setting. However in countries where red meat contamination is of more concern and should the incidence of carcase contamination with pathogens such as *E. coli* O157 increase locally from the current low level, the use of LPS could have a role in these situations. However it should be noted that the effect of LPS against this particular pathogen is not of a great magnitude (less than 1 log).

Further research would allow a better understanding of the antimicrobial effects and benefits of LPS.

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Attachment 4

TOXICOLOGICAL ASSESSMENT

APPLICATION A404 – LACTOPEROXIDASE SYSTEM

Introduction

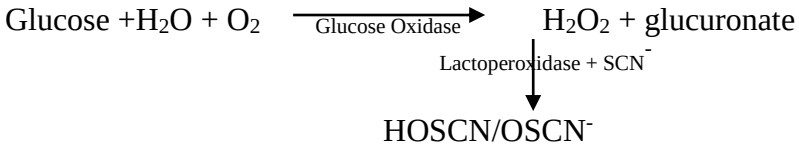
The toxicological assessment of Application A404 to approve the use of the lactoperoxidase system (LPS) for antimicrobial action on meat involves the consideration of three separate components:

(a) **the lactoperoxidase enzyme** (extracted from bovine milk) – at levels between 1 and 20 mg/kg of meat

(b) **a source of thiocyanate ions** (sodium thiocyanate or potassium thiocyanate) – at levels between 5 and 40 mg/kg of meat (as the thiocyanate ion SCN^-)

(c) **a source of hydrogen peroxide** (*in situ* generation using glucose oxidase) – at levels between 5 and 50 mg/kg of meat.

The mode of action of the lactoperoxidase system relies on the production of short-lived intermediary oxidation products of the thiocyanate ion, principally the hypothiocyanate ion, though sulfurdicyanide and cyanosulphurous acid have also been suggested (Oram & Reiter, 1966, Hogg & Jago, 1970). These ions in turn react with the bacterial cytoplasmic membranes, as well as impair the function of metabolic enzymes, hence their anti-microbial effect (Mickelson, 1977 and Reiter & Marshall, 1979). The overall reaction is as follows:



The lactoperoxidase enzyme

The lactoperoxidase enzyme is present naturally in human and cow's milk. Similar enzymes are also present in salivary, thyroid and lacrimal glands. The levels in milk vary, but range around 30 mg/litre, a concentration similar to or slightly higher than that proposed for use in the LPS. The lactoperoxidase used in the system is identical to that found in milk, and indeed is extracted from milk (skimmed, refrigerated and pasteurised), using an ion-exchange column. Lactoperoxidase is not considered to pose any toxicological risk however the enzyme preparation may contain up to 30% milk protein which may be allergenic for sensitive individuals.

Thiocyanate ion source (sodium/potassium thiocyanate)

The thiocyanate ion (SCN^-) is widely distributed in animal tissues and secretions, including the mammary, salivary and thyroid glands, and in the stomach and kidneys. It also occurs in

several food groups including brassicae (where it is of glucosinolate origin) and legumes (where it is of glycoside origin).

The levels in these foods are higher than those proposed for use in the lactoperoxidase system (5-40 ppm), with levels in brassicae reaching up to 100 ppm. In human body fluids levels typically range from 10 to 200 ppm (Reiter & Harnulv, 1984, Farrag & Marth, 1992), and in bovine milk from 1 to 10 ppm (Reiter & Harnulv, 1984).

The thiocyanate ion has been shown to have toxic effects at high levels, with excessive intake interfering with iodine metabolism, and hence thyroid function. However, in studies of SCN^- in milk, effects on iodine uptake in man were only seen with levels of 200-400 ppm, a level far higher than would result from the use of the lactoperoxidase system (Vilkki & Piironen, 1962). The LD50 dose of orally administered sodium thiocyanate in rats, a measure of acute toxicity, is reported to be 764 mg/kg (IDF, 1988).

Information on the reproductive and developmental toxicity of sodium and potassium thiocyanate is limited, especially at the proposed levels of use. Data has been obtained from studies in rats and dogs, at levels proposed for earlier use in hypertension treatment. No adverse effects were found on the growth rate of rats treated with 100 mg/kg for 12 days, although dogs were affected at this level (Anderson & Chen, 1940). The use of SCN- for medicinal purposes has since been stopped due to the narrow margin between therapeutic and toxic concentrations, and the variability in sensitivity of individuals.

No data were available on the genotoxicity of sodium thiocyanate.

Overall, the risk to humans of exposure to the proposed levels of sodium or potassium thiocyanate is very small. At the highest rate of thiocyanate addition, even with zero utilisation of thiocyanate in the process, an adult consuming 100 grams of treated food per day would consume only 4 mg of thiocyanate. For a 60 kg human this equates to a rate of consumption of 0.07 mg per kg per day, over two orders of magnitude less than that used in the rat studies.

Hydrogen peroxide (H_2O_2) source (glucose oxidase)

The enzyme glucose oxidase is currently listed in Standard A16 as an approved processing aid, when sourced from the organism *Aspergillus niger*. To this end, it has already undergone toxicological assessment and approval.
The hydrogen peroxide produced in the presence of glucose oxidase is another issue for toxicological assessment. Although not added to the system, hydrogen peroxide is produced as an intermediate in the reaction of glucose and oxygen. This is dealt with in the following section – Intermediary Products.

Intermediary Products

Hydrogen peroxide

The toxicology of H_2O_2 has been reviewed in the Department of Health and Family services in 1993, has also been evaluated in an IARC monograph in 1985 and by ECETOC (Joint Assessment of Commodity Chemicals no. 22, January, 1993). The US Environmental Protection Agency, after a full toxicological assessment, has established an exemption from the requirement of a tolerance for residues of the biochemical H_2O_2 on all food commodities when used as an algaecide, fungicide and bactericide at the rate of 1% H_2O_2 per application on growing crops and post harvest potatoes (vol 64, no 118, June 1999).

Exogenous H_2O_2 decomposes to oxygen and water on contact with tissues, thus limiting absorption of the intact molecule. Absorbed H_2O_2 undergoes rapid spontaneous or enzyme catalysed decomposition in the epidermis or mucous membranes. Endogenous H_2O_2 formed as a product of anaerobic metabolism is metabolised further by catalase, mainly in peroxisomes, and by glutathione peroxidase in cytosol and mitochondria.

Although hydrogen peroxide is generated by the oxidation of glucose that occurs naturally during the action of glucose oxidase, it is generally assumed not to be present in milk. This is because H_2O_2 is rapidly reduced during the enzymatic oxidation of thiocyanate to produce the hypothiocyanate ion, producing water. The theoretical potential presence of H_2O_2 is therefore not considered a toxicological risk. Even if small quantities were present, which as discussed is unlikely, hydrogen peroxide is in fact approved for use as a bleaching agent in Standard A16, at a level of 5 mg/kg. To this end, levels of 5 mg/kg are considered to pose no significant risk.

Hypothiocyanate ion

As mentioned, this is the principle active agent in the LPS. Like hydrogen peroxide, it is only ever present at very low levels, is very short-lived, and breaks down to form harmless by-products. It is also found naturally in human saliva (Thomas et al, 1980). Its presence as an intermediary product is therefore not considered to pose a toxicological risk.

Residual protein

The applicant notes that there is a possibility of residual protein present on the meat, as a result of the treatment. However, the maximal amount of protein that could be present is 70 mg per kg meat. If cooked, this protein would be denatured, though it would remain if the meat were served uncooked, for example, raw beef (carpaccio).

Purity of system components

Lactoperoxidase

Historically, enzymes used in food processing have been found to be non-toxic, and the main toxicological consideration is in relation to possible contaminants. Since lactoperoxidase is not listed in the Food Chemicals Codex, no specification exists for levels of impurities. According to the applicant, lactoperoxidase, as supplied by Tatua Co-operative Dairy Co. Ltd., has a purity of over 75%, based on the actual lactoperoxidase content of the commercial product. Impurities present are milk salts and milk proteins, neither of which should pose any toxicological risk, although the possibility that their presence may cause problems for milk-allergic individuals. Lactoperoxidase used for the preservation of milk is recommended by the International Dairy Federation to have a purity of 98-99%.

Sodium thiocyanate

The principle impurities of concern as specified by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) are heavy metals, sulphates and sulphides. The purity requirements with respect to these compounds are shown in Table 1. According to the applicant the sodium thiocyanate intended for use in the lactoperoxidase system meets these specifications.

Table 1. JECFA requirements for sodium thiocyanate purity

Criteria	Specification
Heavy Metals	Less than 2 ppm
Sulphates	Less than 50 ppm
Sulphides	Less than 10 ppm

Potassium thiocyanate

Potassium thiocyanate is not listed in the Food Chemicals Codex or the British Pharmacopoeia. However, according to the applicant, the potassium thiocyanate intended for use in the lactoperoxidase system meets the same purity specification as the sodium thiocyanate, as outlined above.

Glucose oxidase

Since glucose oxidase is already included in the list of approved processing aids in Standard A16, no purity profile is required.

Summary and conclusions

The safety of the lactoperoxidase system has been assessed by several authors with respect to its use in the preservation of milk (Reiter & Harnulv, 1984, Farrag & Marth, 1992). As noted in the toxicological assessment described above, there are several features that support its safe use:
The occurrence of all three components in human and animal systems;
The high levels of thiocyanate present naturally in human saliva and gastric juice;
The presence of the intermediate compound OSCN- in human saliva;
The short-lived nature of intermediate compounds;
The selective damage to the bacterial cytoplasmic membrane but not to mammalian cell membranes.

As well as these features, toxicological studies in rats have shown that toxic effects are only seen at levels far higher than those proposed for use. From a toxicological point of view, therefore, it is concluded that at the levels of use proposed in the application neither the components of the lactoperoxidase system, nor the intermediary products, pose a significant risk to human health, provided consumers allergic to milk protein are aware of its presence.

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