Bureau Nieuwe Voedingsmiddelen
Novel Foods Unit

Extract van gefermenteerde sojabonen (Touchi)

Extract of fermented soybeans (Touchi)

Tweede beoordeling van de veiligheid voor de consument, volgens de Europese verordening 258/97 betreffende nieuwe voedingsmiddelen en nieuwe voedselingrediënten

Second opinion regarding consumer safety, in accordance with European Regulation 258/97 concerning novel foods and novel food ingredients

aan/to:

de Minister van Volksgezondheid, Welzijn en Sport
the Minister of Health, Welfare and Sport

Nr. 2009-01 BNV, Den Haag, 17 april 2009
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Beoordeling

Inleiding

Aan de orde is een tweede beoordeling volgens de Europese Verordening 258/97, over het gebruik als nieuw voedselingrediënt van een extract van gefermenteerde sojabonen.

De aanvraag is ingediend door Cantox voor CBC Co. Ltd, uit Japan. Het product uit deze aanvraag is een eiwitrijk poeder, dat wordt aangeduid als Touchi extract (TE). Het wordt geproduceerd uit gele sojabonen (*Glycine max*) die zwart kleuren door een fermentatieproces met behulp van *Aspergillus oryzae.* Deze schimmel wordt al jarenlang gebruikt voor de productie van sojasaus, miso en sake.

De fabrikant zal het nieuwe product op de markt brengen als voedingssupplement in de vorm van capsules en in zakjes voor de bereiding van een drank, te consumeren als thee of soep. De beoogde daginname van 4,5 g TE komt overeen met een gebruikelijke daginname van 15 g sojasaus. De marketing van dit product zal vooral gericht zijn op consumenten die de koolhydraatafbraak willen remmen (afslankdieet). In de EU werd TE niet toegepast in gewone voedingsmiddelen voor 15 mei 1997 en daarom is een veiligheidsbeoordeling als nieuw voedingsmiddel vereist. In het kader van de desbetreffende Europese toelatingsprocedure is deze tweede beoordeling uitgevoerd door het Bureau Nieuwe Voedingsmiddelen van het College ter Beoordeling van Geneesmiddelen. Het bureau heeft hiervoor de Commissie Veiligheidsbeoordeling Nieuwe Voedingsmiddelen geraadpleegd, hierna genoemd ‘de Commissie VNV’.

Eerste beoordeling

De eerste beoordeling van de aanvraag voor markttoelating is verricht in het Verenigd Koninkrijk door de *Advisory Committee on Novel Foods and Processes (ACNFP*).

De ACNFP concludeert dat de voorgestelde toepassingen voor TE acceptabel zijn en heeft geen bezwaar tegen markttoelating. De ACNFP noemt het feit dat producten met gefermenteerde sojabonen wereldwijd al eeuwenlang worden geconsumeerd in de vorm van sojasaus, miso en sake. TE wordt op een vergelijkbare wijze bereid als sojasaus. Daarom wordt niet verwacht dat na inname van TE nadelige gezondheidseffecten zullen ontstaan, mits TE wordt geproduceerd conform de specificatie en productieparameters en voor de etikettering de algemeen geldende richtlijnen worden gevolgd. Na navraag door de ACNFP om de term ‘nutritional support ’ te verduidelijken liet de aanvrager weten dat deze term uitsluitend is toegevoegd om aan te geven dat het de bedoeling is om TE alleen tijdens de maaltijd te gebruiken.

De aanvrager zal voor EU-consumenten een meldingssysteem voor bijwerkingen opstellen en de ACNFP vraagt naar de resultaten na 18 maanden van een post-marketing onderzoek.

Bevindingen van de Commissie VNV

De Commissie VNV heeft geen bezwaar tegen de toelating van TE in de EU en is het eens met de positieve beoordeling door de ACNFP, maar heeft daarbij wel enkele opmerkingen. Zij heeft haar oordeel gebaseerd op de informatie in het dossier, waarvan de samenvatting is opgenomen als bijlage A, de eerste beoordeling door de ACNFP, toegevoegd als bijlage B, en andere bronnen.

Het uitgangsmateriaal voor TE is een varieteit van de sojaboon (*Glycine max*), ook wel 'small yellow bean' genoemd. Deze sojaboon wordt op grote schaal geteeld en al jarenlang geconsumeerd in de provincie Sichuan in China.
In het dossier staat het productieproces van TE als volgt omschreven: na de oogst worden de sojabonen gewassen, gezuiverd van vreemd materiaal, en vervolgens gestoomd en gefermenteerd in een aerobische omgeving met behulp van de schimmel *Aspergillus oryzae* tot 'fermented black beans'. Deze fermentatiemethode wordt reeds honderden jaren op industriële schaal toegepast, zowel op soja als rijst of andere granen. Het gefermenteerd product wordt vervolgens vermalen en met heet water geëxtraheerd. Na een aantal zuiveringstappen (sterilisatie en filtratie) ontstaat het eindproduct, een eiwitrijk poeder, dat Touchi Extract (TE) wordt genoemd.

De productspecificatie bevat parameters voor uiterlijk, smaak en eiwitgehalte (> 55% w/w) en limieten voor potentiële verontreinigingen (zware metalen, dioxines, PCB's, PAK's en microbiële verontreinigingen). Het gehalte aan koolhydraten bedraagt 25-30% maar is niet opgenomen in de productspecificatie. In het dossier worden analyseresultaten van drie batches gepresenteerd en deze voldoen aan de productspecificatie. Dit geldt ook voor de resultaten van de stabiliteitsanalyse.

De ACNFP is van mening dat de kwaliteit van het product gewaarborgd is, mede ondersteund door de HACCP- en GMP-certificering. De Commissie VNV is het daarmee eens.

In de gebieden van herkomst is al honderden jaren ervaring met de consumptie van met *Aspergillus oryzae* gefermenteerde sojabonen. Daarvan afgeleide producten als sojasaus, miso en sake kennen een eeuwenlange traditie van gebruik in China en Japan, maar worden ook in Europa al jarenlang gebruikt in Chinese gerechten.

De ACNFP heeft aanvullende gegevens opgevraagd om de overeenkomst in samenstelling tussen TE en een traditioneel product van gefermenteerde sojabonen nader te onderbouwen. De verstrekte HPLC-analyseresultaten geven volgens de ACNFP voldoende onderbouwing dat qua eiwitsamenstelling TE vergelijkbaar is met gefermenteerde bonen en de Commissie VNV sluit zich daarbij aan.

Volgens de aanvrager kan TE, mits geconsumeerd tijdens de maaltijd, de afbraak van koolhydraten in de dunne darm remmen. Dit wordt verklaard door een mogelijke remmende werking van TE op de activiteit van het enzym α-glucosidase, waardoor de omzetting van koolhydraten in glucose en fructose afneemt. Volgens de aanvrager zou dit effect mogelijk een ondersteunende rol kunnen spelen bij gewichtbeheersing. Met betrekking tot de uiteenlopende gemeten waarden voor enzymremming merkt de ACNFP op dat dit effect van invloed kan zijn op de werkzaamheid en niet relevant is bij deze veiligheidsbeoordeling.

De commissie VNV merkt op dat in verschillende landen, waaronder Nederland, α-glucosidase remmende geneesmiddelen op de markt zijn ter stabilisering van het bloedsuikergehalte bij diabetici. De Commissie VNV concludeert met de ACNFP dat de α-glucosidase remmende activiteit in TE geen gezondheidsrisico vormt met betrekking tot het bloedsuikergehalte en ziet daarin geen probleem bij normaal gebruik.

Volgens het dossier zal TE in voedingssupplementen op de markt komen in de vorm van tabletten of als poeder voor de bereiding van een warme drank (vergelijkbaar met thee of instantsoep) om tijdens de maaltijd te consumeren (dat zal ook op het etiket komen te staan). Als **voedingssupplement in tabletvorm** zal bij normaal gebruik de inname maximaal 4,5 g per dag TE bedragen. Dit komt overeen met de hoeveelheid gefermenteerd bonenextract in een dagportie sojasaus (ca. 15 g).
Om een schatting te maken van de mogelijke inname van TE als **thee/soep**, wordt in het dossier gerekend op basis van consumptiegegevens voor thee en instantsoep. Op basis van innamegegevens van thee en instant-soepproducten is voor TE geschat dat de gemiddelde daginname van TE als soep ongeveer 4,5 g (1 portie) en als thee 13.5 g (3 porties) zal zijn. In absolute zin wordt de hoogste inname bepaald op 40,5 g TE per dag (97.5^{e} percentiel van gebruikers 16-64 jaar), uitgaande van 8 porties thee en 1 portie soep per dag. Aangezien TE bedoeld is om tijdens de maaltijd te gebruiken, acht de aanvrager het niet erg waarschijnlijk dat deze hoge innames in de praktijk zullen optreden.

De Commissie VNV is het eens met de ACNFP en voorziet bij normaal gebruik geen veiligheidsrisico op basis van de voorgestelde hoeveelheid in te nemen TE. Zie ook de NOAEL-gegevens hieronder.

Volgens de aanvrager zijn in Japan, waar TE is toegelaten als Food for Specific Health Use (FOSHU) in overeenkomstige producten voor diabetici geen klachten over bijwerkingen gerapporteerd. Het dossier geeft de karakteristieken weer van een achttal klinische studies, waarin eveneens geen ongewenste effecten zijn gemeld.

*A*CNFP concludeert dat op grond van de gegevens van eerdere blootstelling niet wordt getwijfeld aan de veiligheid, maar vraagt wel om de resultaten van een Post-Marketing Monitoring bij de introductie van TE in de EU.

Volgens het dossier behoort *Aspergillus oryzae* tot de *Aspergillus flavus* groep en heeft zich na een jarenlang selectieproces ontwikkeld tot aangepaste stammen.
Voor nadere informatie over deze schimmel heeft de Commissie VNV ook twee andere bronnen geraadpleegd. De European Food and Safety Agency (EFSA) wil aan *A. oryzae* geen ‘QPS-standaard’ (Qualified Presumption of Safety) verbinden, omdat er nog onvoldoende kennis is over het ontstaan en lange termijn-effecten van mycotoxins, cyclopiazonzuur en β-nitropropionzuur en er bovendien geen erkende identificatiemethode is voor *A. oryzae* (EFSA, 2007). De Environmental Protection Agency (EPA) acht het risico op toxische stoffen laag, door een goed gecontroleerd fermentatieproces en het feit dat geen meldingen zijn gedaan van aflatoxineproductie (EPA, 1997).

De ACNFP is van mening dat door de condities tijdens het fermentatieproces geen secundaire metabolieten zullen ontstaan. De Commissie deelt deze visie.

Uit een door de aanvrager uitgevoerde reeks toxiciteitstudies, concludeert deze dat TE een lage acute orale toxiciteit bezit (LD50 > 5000 mg/kg). In een 28 dagen-onderzoek met ratten zijn geen negatieve aan de inname gerelateerde effecten gevonden. Uit dit onderzoek is voor TE een NOAEL-waarde bepaald van 2.500 mg/kg, wat overeenkomt met 150 g/dag. Daarvan uitgaande is bij normaal gebruik zoals bedoeld door de aanvrager (4,5 g/dag) de veiligheidsmarge 33. Bij een gemiddeld gebruik van 18 g TE uit gecombineerd gebruik van thee en soep samen (4 porties per dag) bedraagt deze 8 en bij de hoogst geschatte inname van 40,5 g/dag is sprake van een factor 3,7.
Het dossier meldt dat geen studies met betrekking tot reproductie en ontwikkeling en chronische toxiciteit voorhanden zijn in de gepubliceerde wetenschappelijke literatuur. Op basis van de resultaten van een 'reverse mutation assay' en een *in vivo* micronucleus test concludeert de aanvrager dat TE niet mutageen, noch genotoxisch is.
De aanvrager heeft in het dossier pre-klinisch onderzoek opgenomen, waaruit zou blijken dat TE geen effect heeft op het centraal zenuwstelsel, cardiovasculair systeem en GI-systeem. Ook werden geen ongunstige metabolische effecten, aanwijzingen voor allergie of ontstekingsreacties waargenomen. Uit farmacologische dierproeven kwamen geen bijwerkingen naar voren. Uit klinisch onderzoek (8 studies) met gezonde vrijwilligers en diabetici werden geen bijwerkingen gerapporteerd.

Wel merkt de Commissie VNV daarbij op dat in de meerderheid van deze onderzoeken herhaalde doseringen werden gebruikt van 1-3 g/dag, dus lager dan de in het dossier beoogde 4.5 g/dag.

Conclusie

De Commissie is het eens met de ACNFP dat de aanvrager het voldoende aannemelijk heeft gemaakt met het totaal aan gegevens, dat er geen schadelijke gezondheidseffecten zijn te verwachten voor de Europese consument.

De Commissie VNV deelt de conclusie van de eerste beoordelaar dat voor TE net als voor alle sojabonen bevattende producten etikettering verplicht zal zijn overeenkomstig de bepalingen van Richtlijn 2003/89/EC.

Assessment

Introduction

This is a second opinion in accordance with European Regulation 258/97 concerning the use of an extract of fermented soybeans as a novel food ingredient.

The application has been submitted by Cantox on behalf of CBC Co. Ltd in Japan. The product to which the application relates is a protein-rich powder referred to as Touchi extract (TE). It is produced from yellow soybeans (*Glycine max*), which turn black as a result of fermentation using *Aspergillus oryzae*. This is a fungus that has been used for many years to produce soy sauce, miso and sake.

The manufacturer intends to market the new product as a food supplement in the form of capsules and in sachets for the preparation of a drink that can be consumed as tea or soup. The proposed daily intake of 4.5 g of TE equates to a normal daily intake of 15 g of soy sauce. The product will be targeted mainly at consumers wishing to inhibit the digestion of carbohydrates (for a slimming diet). TE has not been used in normal foods in the EU prior to 15 May 1997 and therefore requires a safety assessment as a novel food. This second assessment has been carried out under the relevant European approval procedure by the Novel Foods Unit of the Medicines Evaluation Board. The Unit has consulted the Committee on the Safety Assessment of Novel Foods (hereinafter referred to as the VNV Committee) on the subject.

Initial assessment

The initial assessment of the application for market approval was carried out in the United Kingdom by the Advisory Committee on Novel Foods and Processes (ACNFP).

The ACNFP concluded that the proposed uses of TE are acceptable and has no objection to its approval, noting that products containing fermented soybeans have been consumed for centuries all over the world in the form of soy sauce, miso and sake. TE is prepared in a similar way to soy sauce, so the ingestion of TE is not expected to have any adverse health effects, provided it is produced in accordance with the specification and production parameters and the general labelling guidelines are observed. Asked by the ACNFP to clarify the term ‘nutritional support’, the applicant explained that this was added purely to indicate that TE is intended only for consumption during a meal.

The applicant intends to set up a side-effects notification system for EU consumers, and the ACNFP has asked to be furnished with the results after 18 months of post-market monitoring.

Findings of the VNV Committee

The VNV Comittee has no objection to the approval of TE in the EU and agrees with the ACNFP's positive assessment, albeit with some comments. It bases its opinion on the information in the dossier (which is summarized in Annex A), the initial assessment by the ACNFP (attached as Annex B), and other sources.

The raw material from which TE is made is a variety of soybean (*Glycine max*) also known as the 'small yellow bean'. This is grown on a large scale in the Sichuan province of China, where it has been consumed for many years.

The TE production process is described as follows in the dossier: once harvested, the soybeans are washed and screened for foreign material, then steamed and fermented in an aerobic environment using the fungus *Aspergillus oryzae* to produce 'fermented black beans'. This fermentation process has been used on an industrial scale for centuries, applied to rice and other cereals as well as soya. The fermented product is then milled and extracted using hot water. After undergoing various purification processes (sterilization and filtration) the end-product is produced, a protein-rich powder referred to as Touchi extract (TE).

The product specification sets out parameters regarding appearance, taste and protein content (>55% w/w) and limits for potential contaminants (heavy metals, dioxins, PCBs, PAHs and microbial contaminants). The carbohydrate content is 25-30%, but this is not stated in the product specification. The dossier sets out the results of analysing three batches: these complied with the product specification, as did the results of the stability analysis.

The ACNFP's opinion is that the quality of the product is assured, backed up by HACCP and GMP certification, and the VNV Committee concurs.

In the regions of origin, experience of consuming soybeans fermented using *Aspergillus oryzae* goes back centuries. Products derived from this process, such as soy sauce, miso and sake, have a centuries-old tradition of use in China and Japan, and they have been used for many years in Europe in Chinese dishes.

The ACNFP has asked for additional data to establish the similarity in composition between TE and a traditional product made from fermented soybeans. The ACNFP considers that the results of HPLC analysis supplied provide sufficient evidence that TE is comparable to fermented beans in terms of protein composition, and the VNV Committee concurs.

According to the applicant, TE, consumed during a meal, can inhibit the digestion of carbohydrates in the small intestine. The explanation for this is that TE may have an inhibiting effect on the activity of the α-glucosidase enzyme, thus limiting the conversion of carbohydrates into glucose and fructose. According to the applicant, this effect could play a supporting role in weight control. As regards the differing values found for enzyme inhibition, the ACNFP points out that this could affect the product's efficacy; it is not relevant to this safety assessment.

The VNV Committee notes that α-glucosidase-inhibiting drugs to stabilize blood sugar levels in diabetics are on the market in various countries, including the Netherlands. The VNV Committee agrees with the ACNFP that TE's α-glucosidase-inhibiting activity does not

constitute a health risk in relation to blood sugar levels and does not envisage this causing any problems with normal use.

According to the dossier, TE will be marketed in food supplements in the form of tablets or as a powder for the preparation of a hot drink (comparable to tea or instant soup) for consumption during a meal (and this will be stated on the label).
As a **food supplement in tablet form**, the maximum daily intake of TE in normal use will be 4.5 g, which equates to the amount of fermented bean extract in a daily portion of soy sauce (about 15 g).

To estimate the potential intake of TE as **tea** or **soup** the dossier bases its calculations on the consumption data for tea and instant soup. Based on intake data for tea and instant soup products it estimates an average daily intake of TE as soup of 4.5 g (1 portion) and as tea of 13.5 g (3 portions). In absolute terms the maximum intake of TE is estimated at 40.5 g per day (97.5^{th} percentile of users aged 16-64 years), assuming 8 portions of tea and 1 portion of soup per day. As TE is intended for consumption during a meal, the applicant does not think that such high intakes are likely to occur in practice.

The VNV Committee agrees with the ACNFP and does not envisage any safety risk based on the proposed amount of TE being ingested in normal use (see also the NOAEL data below).

According to the applicant, in Japan, where TE is approved as a Food for Specific Health Use (FOSHU) in similar products for diabetics, no complaints of side-effects have been reported. The dossier describes a number of clinical studies in which no adverse effects were found.

The ACNFP concluded that there is no doubt as to the product's safety based on information on previous exposure, but it has asked to be furnished with the results of post-market monitoring if TE is introduced in the EU.

According to the dossier, *Aspergillus oryzae* is a member of the *Aspergillus flavus* group and has undergone years of selection to produce suitable strains.
In order to obtain further information on this fungus the VNV Committee has also consulted two other sources. The European Food and Safety Agency (EFSA) is unwilling to confer 'QPS status' (Qualified Presumption of Safety) on *A. oryzae*, since there is as yet insufficient knowledge on the development and long-term effects of mycotoxins, cyclopiazonic acid and β-nitropropionic acid, nor is there a recognized method for identifying *A. oryzae* (EFSA, 2007). The Environmental Protection Agency (EPA) considers the risk of toxic substances to be low, as the fermentation process is properly controlled and there have been no reports of aflatoxin production (EPA, 1997).

The ACNFP considers that no secondary metabolites will develop as a result of the conditions during the fermentation process, and the VNV Committee shares this view.

The applicant concludes, from a series of toxicity studies it has carried out, that TE has a low acute oral toxicity (LD50 >5000 mg/kg). No negative effects related to ingestion were found in a 28-day study in rats. An NOAEL value of 2,500 mg/kg was found for TE in this study, corresponding to 150 g per day. On this basis, in normal use as defined by the applicant (4.5 g per day) the safety margin is 33. With an average consumption of 18 g of TE

from the combined consumption of tea and soup (4 portions per day) the figure is 8, and with the maximum estimated intake of 40.5 g per day 3.7.

The dossier states that there are no studies in the published scientific literature on reproduction, development or chronic toxicity. Based on the results of a reverse mutation assay and an *in vivo* micronucleus test the applicant concludes that TE is neither mutagenic nor genotoxic.

The applicant includes pre-clinical trial data in the dossier which supposedly indicates that TE has no effect on the central nervous system, cardiovascular system or GI system; nor have any adverse metabolic effects, indications of allergy or inflammatory reactions been observed. Pharmacological experiments on animals have not shown any side-effects. No side-effects have been reported from clinical trials (8 studies) with healthy volunteers and diabetics.

The VNV Committee would point out, however, that these studies used repeated dosages of 1-3 g per day, i.e. lower than the proposed daily intake of 4.5 g in the dossier.

Conclusion

The VNV Committee agrees with the ACNFP that the applicant has adequately demonstrated with the data as a whole that there are not likely to be any harmful health effects for European consumers.

The VNV Committee shares the conclusion of the first assessor that labelling will be required for TE, as for all products containing soybeans, in accordance with Directive 2003/89/EC.

Referenties/References

- Application dossier of CBC Co., Ltd 'Application for the Approval of Touchi Extract under Regulation (EC) No. 258/97 of the European Parliament and of the Council of 27 January 1997 concerning Novel Food Ingredients'.

- Advisory Committee on Novel Foods and Processes (ACNFP). Opinion on an Application under the Novel Foods regulation for Touchi Extract derived from fermentation of soya bean by Aspergillus oryzae as a Food Ingredient (February 2009).

- European Food Safety Agency (EFSA). Qualified Presumption of Safety, The EFSA Journal (2007) 587.

- Environmental Protection Agency (EPA). Final Risk Assessment of Aspergillus oryzae. EPA (US) 1997.

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A Samenvatting van het dossier / Summary of the dossier

APPLICATION FOR THE APPROVAL OF TOUCHI EXTRACT UNDER REGULATION (EC) NO 258/97 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL OF 27 JANUARY 1997 CONCERNING NOVEL FOODS AND NOVEL FOOD INGREDIENTS

SUMMARY

NON-CONFIDENTIAL

CBC Co., Ltd

July 3, 2008

APPLICATION FOR THE APPROVAL OF TOUCHI EXTRACT UNDER REGULATION (EC) NO 258/97 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL OF 27 JANUARY 1997 CONCERNING NOVEL FOODS AND NOVEL FOOD INGREDIENTS

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INTRODUCTION

CBC Co., Ltd. wishes to market Touchi extract (TE) a protein-rich product in powder form obtained by aqueous extraction of small soybeans that have been fermented using the fungus *Aspergillus Oryzae* (also known as “salted black beans”) for use in food supplement products in the European Union.

Fermented black beans have been widely used in China for the past 1000 years as a traditional seasoning and people are familiar with black beans as a protein source that can

be preserved by drying without damage to the nutrient content. In Europe, fermented black beans have also been consumed for many years in Chinese dishes containing “black bean sauce” or involving “black bean paste” as an ingredient, and extensive discussions with the UK Novel Foods Competent Authority have indicated that the use of Touchi extract would not be considered novel for flavouring or seasoning. However, CBC Co., Ltd. to market this product for use in food supplement products in the European Union, Touchi extract would be considered “novel” and fall under category (e) of Article 1(2) of Regulation (EC) No 258/97 (European Parliament and the Council of the European Union, 1997) (foods and food ingredients consisting of or isolated from plants and food ingredients isolated from animals, except for foods and food ingredients obtained by traditional propagating or breeding practises.

The application for placing Touchi extract on the market as a novel food or novel food ingredient is required follow to the European Commission’s Scientific Committee on Food (SCF) Recommendation 97/618/EC (Commission of the European Communities, 1997). Under Section 4 of this Recommendation, pertaining to the scientific classification of novel foods for the assessment of wholesomeness, Touchi extract would be considered as “Class 2 Complex Novel Food from non-GM sources: 2.1 the source of the novel food has a history of use in the Community”. Unlike the well-known large soybean, the small soybean has not been genetically modified and therefore, Touchi extract is derived from a non-GM source.

I SPECIFICATION OF THE NOVEL FOOD

The tradename “Touchi extract” will be used for marketing the food ingredient to food supplement manufacturers. The proposed name for labelling purposes on final food supplement products as presented to the consumer will be Touchi extract” or “Fermented black bean extract” (where “*made from soya” may be used as a footnote). Analytical data has been provided to show that Touchi extract does not contain any detectable potentially toxic constituents or external contaminants that might be associated with a fermented product derived an oilseed such as the small soybean (Commission of the European Communities, 2006b). Specifically, analytical results have been presented for pesticides, heavy metals, dioxins and dioxin-like PCBs, polycyclic aromatic hydrocarbons, mycotoxins and 3-monochloropropane-1,2-diol. Essentially, Touchi extract is a concentrated form of the protein fraction of fermented black beans. The specification of the material is well-defined (Table I-1) and analysis for three non consecutive production batches provided to show that the product can be manufactured consistently in accordance with clearly defined chemical and physical parameters. It is well established that fermented black beans exhibit alpha-glucosidase inhibitory action and data has been provided to show that on extraction this activity is retained. The stability of the product both in the bulk form and in typical product formulations has been established.

Table I-1 Proposed Specification for Touchi Extract		
Parameter	**Specification**	**Analytical Method**
Characteristics		
Appearance	Light brown powder	Visual
Taste	'Pleasant"	Taste
Fat	Max. 1%	Soxhlet extraction
Protein	Min. 55%	Kjeldahl method, AOAC 981
Water	Max. 7%	AOAC 925.1
α-glucosidase inhibitor activity	IC_{50} min. 0.025	Enzyme assay[1]
Contaminants		
Arsenic	Max. 10 µg/kg	Atomic Absorption Spectroscopy
Aflatoxins	Max. 5 µg/kg	HPLC
3-MCPD	Max. 50 µg/kg	GCMS
Total heavy metals (expressed as lead)	Max. 20 µ/kg	Na_2S Colorimetric method
Microbiological Requirements		
Total bacteria count	≤ 1000 cfu/g	USP23
Total mould and yeast count	≤ 300 cfu/g	USP23
Escherichia coli	Negative /g	USP23

[1]Modified method of Miwa *et al*., *Chem Pharm. Bull*., 34:838, 1986

II EFFECT OF THE PRODUCTION PROCESS APPLIED TO THE NOVEL FOOD

The fermentation of the small soybean is achieved in an aerobic environment using the fungus *Aspergillus oryzae*. The resulting fermented black beans are then extracted with water and followed by spray drying to yield Touchi extract as a brown powder. No chemical modification of the fermented black beans occurs and conventional extraction procedures are employed. As an indication of scale, one tablespoon (15 g) of fermented black beans (the amount typically provided in one serving of a dish using black bean sauce) corresponds to 4.5 g of Touchi extract, the daily serving proposed herein. The process is performed in accordance with the principles of Hazards Analysis and Critical Control Points (HACCP) and Good Manufacturing Practise (GMP). The inclusion of a pasteurisation stage and the maximum 7% limit on water content in the product specification both ensure that microbial growth is minimised. Due to the nature of the processing route, if a product fails to meet specification it cannot be reprocessed easily and is therefore discarded.

III HISTORY OF THE ORGANISM USED AS THE SOURCE OF THE NOVEL FOOD

The raw material used to prepare Touchi extract is the small soybean (*Glycine max*.) which has been extensively used in the Sichuan province of China for centuries where it is know as the small yellow bean. The main feature of soybeans that has led to their extensive use in foodstuffs is the high level of protein, but there is also a large market for soybean oil. Fermentation is performed using *Aspergillus oryzae* which has been used for hundreds of years in the production of soy sauce, miso, and sake (Wood, 1977).

The small soybean plant is a species of legume and the soybeans obtained are considered oilseeds. Unlike the well-known large soybean, the small soybean has not been genetically modified and therefore, Touchi extract is derived from a non-GM source.

IX ANTICIPATED INTAKE/EXTENT OF USE OF THE NOVEL FOOD

It is anticipated that Touchi extract is consumed as a nutritional support during a meal in order to hinder the digestion of carbohydrates in the small intestinal tract in an analogous way to food ingredients such as resistant starch or “starch blockers” which are currently on the market in the EU. Such a property may help people dieting feel less hungry for longer after a meal, similar in principle to the purpose of so-called “low glycaemic” foods.

Touchi extract will only be marketed in supplement form either as capsules or in sachet form (*e.g*., tea formulation) to be always taken with a meal at levels that would not exceed 4.5 g per daily serving. Touchi extract will only be added to products as a single dose supplement formulation where presentation to the consumer is controlled and at risk groups protected (*e.g*., diabetics). Risk management measures will be applied under the conditions of Directive 2002/46/EC of the European Parliament and of the Council of 10 June 2002 on the approximation of the laws of the Member States relating to food supplements (European Parliament and the Council, 2002) and also under the conditions of Regulation 1924/2006 of the European Parliament and the Council of 20 December 2006 on nutrition and health claims made on foods (European Parliament and the Council of the European Union, 2007).

Although food supplement products are traditionally available in tablet or capsule form there are examples of these products being provided in sachet form for delivery as stand-alone drinks or for addition to other beverages/foodstuffs currently in the EU. Touchi extract is not proposed for incorporation into conventional foods and therefore a detailed exposure assessment is not warranted. A brief screening of tea and vegetable soup intakes in general for the UK population (EFSA, 2008) however, confirmed there was no safety concern posed even by the highest level consumers of these types of products consuming Touchi extract-containing products as conventional foods and not in accordance with the labelling directions.

X INFORMATION FROM PREVIOUS HUMAN EXPOSURE TO THE NOVEL FOOD OR ITS SOURCE

Fermented black beans have been widely used in China for the past 1000 years as a traditional seasoning and people are familiar with black beans as a protein source that can be preserved on drying without damage to the nutrient content.

Soybeans are a common source of food allergenicity and are included in Annex IIIa of Directive 2002/89/EC regarding the indication of ingredients present in foodstuffs (European Parliament and the Council, 2003). In accordance with this legislation, all food supplement products containing Touchi extract will clearly be labelled as being made from soya or soybeans.

Touchi extract is currently approved as a Food for Specific Health Use (FOSHU) in Japan. CBC Co. Ltd has not received any complaints to date regarding adverse effects on the consumption of products containing Touchi extract.

XI NUTRITIONAL INFORMATION ON THE NOVEL FOOD

From a nutritional safety perspective Touchi extract can be considered substantially equivalent to fermented black beans and fermented black bean paste which have been on the market in the European Union (EU) for many years. For comparison, the nutrient profiles for 15 g of fermented black beans and 4.5 g of Touchi extract are provided in Table XI-1. The relative amount of each nutrient changes during processing because the water-insoluble fraction is removed (*e.g.*, the fat content), however no chemical modification is involved, therefore, the nature of the nutrients remains the same for the extract and the fermented black beans.

Table XI-1 Comparison of Nutrient Profiles for Fermented Black Beans and the Extract

Constituent	Composition (% w/w total)	
	Fermented Black Beans (Data from Japanese Food Index)	TE (Lot Manufactured 17/3/05)
Protein	18.6	61.2
Fat	8.1	0.2
Carbohydrate	31.5	27.3
Water	24.4	3.5
Ash	17.4	7.8
Total	100	100

Soy proteins are generally regarded as “complete proteins” providing all of the essential amino acids in sufficient quantities to meet human nutritional needs and are typically resistant to denaturing by cooking and other processing methods (FAO, 1991). Soybeans contain a number of isoflavones which are flavonoids with structural similarities to oestrogen.

Touchi extract can inhibit alpha-glucosidase action, thus aiding the body’s self-regulation of this enzyme. As a consequence, consumption of Touchi extract as a food supplement alongside a meal can delay carbohydrate digestion in the small intestinal tract (Clissold and Edwards, 1988; Toeller, 1994; Fujita *et al*., 2005). Undigested carbohydrates or disaccharide are then excreted rather than being absorbed by the body thus potentially offering assistance in weight control regimes. The effect of Touchi extract on carbohydrate digestion is analogous to that observed in a variety of foods including indigestible dextrin, resistant starch and alpha-amylase inhibitors (so-called “starch-blockers”).

XII MICROBIOLOGICAL INFORMATION ON THE NOVEL FOOD

The results of microbiological testing (Table XII-1) confirm the absence of pathogens which as discussed, earlier, is expected from a product which has undergone a sterilisation process and has a low water content (limit 7%).

Table XII-1 Microbiological Analysis for Touchi Extract

Contaminant	Microbiological Growth Results		
	Lot 1	Lot 2	Lot 3
Total bacteria count	Less than 1000 cfu/g	<1000	<1000
Total mould and yeast count	Less than 300 cfu/g	<300	<300
Escherichia coli	Negative /g	Negative	Negative

XIII TOXICOLOGICAL INFORMATION ON THE NOVEL FOOD

The safety of Touchi extract is based principally on its equivalence to fermented black beans which have been consumed since ancient times and is widely available in culinary dished throughout the EU. At the maximum consumption level of 4.5 g per day, Touchi extract is equivalent to approximately a tablespoon (15 g) serving of fermented black beans. The safety of Touchi extract at these levels is further supported by a range of toxicological studies, including a 28-day subchronic study in the rat which established a no-observed-adverse-effect level (NOAEL) of 2500 mg/kg body weight/day (Fujita and Yamagami, 2007) equivalent to 175 g/day for a 70 kg adult equating to a 40-fold safety factor compared to a maximum daily consumption of 4.5 g. Numerous human clinical studies have been conducted in both healthy and diabetic subjects with Touchi extract at levels up to 10 g per daily serving and durations of up to 6 months (see Table XIII-1). The absence of major adverse effects in subjects offers additional evidence for the safety of Touchi extract.

Table XIII-1 Clinical Studies Conducted with Touchi Extract

Reference	Study Design	Subject Population	Dosing Regimen	Endpoints Measured	Results/ Evaluation of Safety Parameters
Safety Data on Healthy Subjects					
Nippon Supplement Inc. Research and Development, 1999a	Safety	3 healthy male subjects	Single bolus dose of 10 g TE	Haemological, biochemical, and urinary parameters, objective and subjective symptoms	No notable abnormalities were observed in any test.
Nippon Supplement Inc. Research and Development, 1999b	Safety	10 healthy male subjects	1 g TE before each meal (3 g/day) for 12 weeks	Haemological, biochemical, and urinary parameters, objective and subjective symptoms	No notable abnormalities were observed in any test.
Hiroyuki *et al.,* 2001	Confirmation of safety	9 healthy males (26-56 years of age)	1 g TE 3 times daily before meals for 12 weeks	Haemological and biochemical parameters, body weight, and body mass index, subjective symptoms	No remarkable changes in haemological, relevant biochemical data, body weights, or body mass indices were observed. Fasting blood glucose and glycated haemoglobin were not altered and there were no reports of gastrointestinal symptoms or any other adverse effects related to ingestion of TE.
Data on Diabetic Subjects					
Hiroyuki *et al.,* 2001	Non-comparative study	18 subjects (male and female) with non-insulin dependent diabetes mellitus	0.3 g TE 3 times daily before each meal for 6 months	Haemological and biochemical parameters, body weights and body mass indices, plasma lipids, and subjective symptoms	Fasting blood glucose and glycated haemoglobin levels were significantly reduced after 6 months of TE ingestion. The extract effectively attenuated the fasting blood glucose levels in 14 subjects (77.8%) and glycated haemoglobin levels in 11 patients (61.1%). A moderate decrease in total cholesterol levels was observed while high-density lipoprotein levels were reportedly moderately increased. Triglyceride levels were significantly decreased after 3 and 6 months of treatment. There were no complaints of gastrointestinal side effects and no remarkable changes in haemological, relevant biochemical parameters, body weights, or body mass indices were observed.

Table XIII-1 Clinical Studies Conducted with Touchi Extract

Reference	Study Design	Subject Population	Dosing Regimen	Endpoints Measured	Results/ Evaluation of Safety Parameters
Fujita *et al.*, 2001a	Effects of Touchi on blood glucose levels after sucrose-loading	8 borderline diabetic subjects 4 diabetic subjects	0.1-10.0 g TE before sucrose loading (75 g) 0.3 g TE before eating 200 g of cooked rice	Blood glucose and insulin levels, haemological and biochemical parameters, subjective side effects	Dose-dependent decreases the glycaemic response were observed. Significant suppression of postprandial blood glucose levels was seen at 60 and 90 minutes after sucrose loading. Area under the curve showed significant antiglycaemic effects at a minimum effective dose of 0.3 g. The postprandial increases in both blood glucose and mean insulin levels observed with TE administration were significantly depressed at 60 and 120 minutes after ingestion compared with levels when no TE was administered. Neither borderline nor diabetic patients complained of any side effects such as abdominal pain, diarrhoea, retching or flatulence after the ingestion of TE. No abnormalities in haemological or biochemical parameters were observed.
Fujita *et al.*, 2001b	Double-blind, placebo-controlled study	36 subjects (15 males, 21 females) with borderline and mild type-2 diabetes	Houji-tea with steamed soybean powder (placebo) or 0.3 g of TE before meals, 3 times per day for 3 months	Haemological and biochemical parameters, subjective symptoms	Glycated haemoglobin (HbA1c) and fasting blood glucose levels were significantly reduced after 2 and 3 months, respectively, with TE supplementation. Triglyceride concentrations also tended to decrease at 2 months post-TE ingestion. No such effects were observed in the placebo group. No other significant changes on haemological or biochemical parameters or body weights and body mass indices were observed, and there were no reports of gastrointestinal side effects or other adverse events. There was no deterioration as assessed by fasting blood glucose and glycated haemoglobin levels after withdrawal of TE.
Fujita *et al.,* 2003	Randomised, double-blind, placebo-controlled	47 borderline and mild type-2 diabetic subjects	0.3 g TE before meals, 3 times daily for 6 months	Blood glucose and insulin levels, haemological and biochemical parameters, subjective side effects	Fasting blood glucose, glycated haemoglobin levels, and triglyceride levels were significantly decreased in the TE-treated group. Total cholesterol levels tended to decrease in subjects consuming TE, however this decrease did not reach statistical significance. The high-density lipoprotein (HDL) content was unchanged in both the placebo and TE-treated groups. No adverse effects on haemological or biochemical parameters were observed. Body weight and BMI values at the completion of the study were comparable to pre-study values. No subjective

Table XIII-1 Clinical Studies Conducted with Touchi Extract

Reference	Study Design	Subject Population	Dosing Regimen	Endpoints Measured	Results/ Evaluation of Safety Parameters
					side effects were reported.
Fujita *et al.*, 2005	Randomised, double-blind, placebo-controlled study	50 nondiabetic, mild and borderline hypertriglcyeridemic subjects	0.3 g of TE in tablet form before meals 3 times daily for 6 months	Haemological and biochemical parameters, subjective symptoms	Four subjects were excluded from the study for reasons unrelated to their health and the administration of TE. Of the 46 subjects that completed the study, 25 received the TE tablets while 21 ingested placebo tablets. After 1 month, the triglyceride level in treated subjects was lower than the initial baseline value. This trend continued as the study progressed, and significant reductions were observed at the 2-, 4-, and 6-month time points compared to baseline. In contrast, the TG level of the placebo group did not change significantly. Significant reductions in mean TG levels were seen in treated subjects compared to placebo after 2, 4, and 6 months. Results were evaluated again by subdividing both the placebo and treatment groups into 2 subgroups each based on initial TG levels (borderline *vs*. mild hypertriglyceridemia). Both borderline and mild hypertriglyceridemics experienced significant reductions in TG levels compared to their respective baseline values after treatment with TE. In subjects with borderline hypertriglyceridemia, significant decreases in TG levels were observed at 2 and 4 months compared to borderline hypertriglyceridemic subjects in the placebo group. Significant decreases in TG levels were seen at 4 and 6 months in treated mild hypertriglyceridemics compared to placebo subjects with mild hypertriglyceridemia. After 1 month, the total fasting blood glucose level in the TE-fed subjects (both mild and borderline hypertriglyceridemic) had decreased, with significant decreases seen at 4 and 6 months. In contrast, the total cholesterol levels of treated and placebo groups did not change markedly, and there were no changes in total HDL levels in treated or placebo groups over the study period. Total body weight and BMI of the treatment and placebo groups did not differ significantly over the study period. The subjects

Table XIII-1 Clinical Studies Conducted with Touchi Extract					
Reference	**Study Design**	**Subject Population**	**Dosing Regimen**	**Endpoints Measured**	**Results/ Evaluation of Safety Parameters**
					reported no side effects such as abdominal distention, abdominal pain, diarrhoea, retching, increased flatulence, or allergic symptoms. No adverse effects were seen on haemological and biochemical parameters.

OVERALL CONCLUSIONS

The safety of Touchi extract is based principally on its equivalence to fermented black beans, which have been consumed since ancient times and are widely available in culinary dishes throughout the European Union. At the maximum consumption level of 4.5 g per day, Touchi extract is equivalent to approximately a 1 tablespoon (15 g) serving of fermented black beans. The solvent used is water and no selective extraction occurs with the result that no chemical modification occurs and the composition is comparable to the traditional counterpart, fermented black beans. Touchi extract has the ability to inhibit the activity of the alpha-glucosidase enzyme so delaying the digestion of carbohydrate in the small intestinal tract following consumption of food. Undigested carbohydrates or disaccharides are then excreted rather than being absorbed by the body thus providing possible assistance in weight control regimes. The approval of Touchi extract is requested only for specific food supplement products (in tablet or sachet form) that would be taken with meals at levels not exceeding 4.5 g/day and as such would have specific risk management and labelling clauses that would prevent the consumption by at risk groups such as diabetics. The safety of Touchi extract at these levels is further supported by a range of toxicological safety studies, including a 28-day sub-chronic study in the rat, which established a NOAEL of 2500 mg/kg body weight/day, equivalent to 150 g/day for a 60 kg adult or to approximately a 33-fold safety factor compared to maximum daily consumption of 4.5 g. Numerous human clinical studies have been conducted in both healthy and diabetic subjects with Touchi extract at levels up to 10 g per daily serving and durations of up to 6 months. The absence of major adverse effects in subjects offers additional evidence for the safety of Touchi extract.

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B Eerste beoordeling / Initial assessment

ADVISORY COMMITTEE ON NOVEL FOODS AND PROCESSES

OPINION ON AN APPLICATION UNDER THE NOVEL FOODS REGULATION FOR TOUCHI EXTRACT DERIVED FROM FERMENTATION OF SOYA BEAN BY *Aspergillus oryzae* AS A FOOD INGREDIENT

Applicant: **CBC Co Ltd**

Responsible Person: **Mr Shinya Miyairi**

EC Classification: **2.1**

Introduction

1. An application was submitted by CBC Co. Ltd for the authorisation of Touchi extract derived from the fermentation of black bean (*Glycine max*) by *Aspergillus oryzae* as a novel food ingredient.
2. Touchi extract is a protein-rich powder obtained by water extraction of small soybeans fermented with *Aspergillus oryzae* and contains an alpha-glucosidase inhibitor. It is intended to be consumed by people who wish to slow the breakdown of carbohydrate following a meal.
3. This application for the authorisation of Touchi extract was prepared pursuant to Commission Recommendation 97/618/EC of 29 July 1997 concerning the scientific aspects and presentation of information necessary to support applications for the placing on the market of novel foods and novel food ingredients. Touchi extract has been classified as a complex novel food from a non-GM source, where the source of the novel food has a history of consumption in the EU (class 2.1).

I. Specification of the novel food

Application dossier, pp 4-12

4. The raw material used to prepare the novel ingredient (NI) is a variety of soybean (*Glycine max*) which has been extensively used in the Sichuan province of China for centuries and is also known as the small yellow bean. Fermentation is performed using *Aspergillus oryzae* which has an established use for many years in the production of soy sauce, miso and sake.

5. The NI is a light brown powder and the applicant has provided the following proposed specification which includes both nutrient and purity limits:

Specification for Touchi Extract	
Parameter	**Specification**
Characteristics	
Appearance	Light brown powder
Taste	'Pleasant"
Fat	Max. 1%
Protein	Min. 55%
Water	Max. 7%
α-Glucosidase inhibitor activity	IC_{50} min. 0.025
Contaminants	
Arsenic	Max. 10 µg/kg
Aflatoxins	Max. 5 µg/kg
3-MCPD	Max. 50 µg/kg
Total heavy metals (expressed as lead)	Max. 20 µg/kg
Microbiological Requirements	
Total bacteria count	≤ 1000 cfu/g
Total mould and yeast count	≤ 300 cfu/g
Escherichia coli	Negative /g

6. The applicant states that no pesticides are used in the production of the NI. Results of a pesticide residue screen show none were present at or above the limits of detection.
7. The applicant has provided an analysis of potentially toxic inherent constituents, external and process contaminants, for three non-consecutive batches of the NI which are representative of the commercial product to be marketed in the EU.
8. Heavy metal analysis show levels of lead and cadmium are within acceptable ranges (0.2 µg/kg limit for lead in cereals, legumes and pulses and for cadmium in soybean, according to EU legislation) and levels for arsenic and mercury are also below the limit of detection.
9. At the proposed level of intake for the NI, the levels of dioxins and dioxin-like PCB's analysed in three batches are considered to fall within acceptable ranges. Levels of Polycyclic Aromatic Hydrocarbons (PAH) are also below detection limits.
10. The applicant's mycotoxin screen indicated that none of the batches of the NI contained aflatoxins B_1, B_2, G_1 and G_2 at the limit of detection (<0.2 µg/kg). The applicant states that as far as it is aware *Aspergillus oryzae* is not a source of ochratoxin A and this statement was supported by an expert at an accredited independent testing laboratory.
11. The level of 3-monochloropropane-1,2-diol (3-MCPD) was determined for three batches of the NI. The levels of 3-MCPD were found to be <30 µg/kg for the three

batches tested and below the EU regulatory requirements (up to 50 µg/kg in dry matter). The applicant highlights that the maximum intake level of up to 4.5 g/day proposed for the NI ensures this product will not contribute significantly to the dietary intake of 3-MCPD.

12. The applicant notes that one portion of a dish using traditional black bean sauce would generally contain 15 g of fermented black beans, which on extraction, is equivalent to 4.5 g of the NI. During extraction of fermented black beans, the water soluble fraction is removed from the insoluble part to give a product that is high in protein but low in fat. The applicant states that, as no chemical modification is involved in the extraction of the water-soluble fraction of fermented black bean, the nature of the nutrients remains identical to the traditional fermented counterparts and therefore the NI is essentially a concentrated form of the protein fraction of fermented black beans. The applicant has provided a summary of the fat and protein content of 3 batches of the NI in Table I.C.2.1-1 of the application dossier.

13. Fermented black beans exhibit alpha-glucosidase inhibitory action and on extraction this activity is retained. The IC_{50} for inhibition of alpha-glucosidase ranged from 0.05 to 0.55 mg/mL in three batches of the NI. The NI was also screened for inhibition of other enzymes but only alpha-glucosidase inhibitory action was observed.

14. The NI is intended to be used as a powder or incorporated into formulations for presentation to the consumer as a food supplement. In order to confirm the stability of the NI the product was monitored as bulk powder, tablet form and tea formulation. The NI was observed to be stable for over 36 months at room temperature based on these studies. In addition, results of accelerated tests at 55°C and tests as a 10% (w/w) solution in water were provided

***Discussion**: The Committee accepted the applicant's proposed specification for the NI and was satisfied with the stability tests. The Committee noted that the wide range of IC_{50} values reported for the NI may affect the efficacy of different batches of the product but not their safety. (Please also refer to paragraph 27 below.)*

II. Effect of the production process applied to the novel food

Application dossier, pp 13-17

15. The applicant provides a basic overview of the fermentation process in the application dossier, noting that this is consistent with the conditions used in the production of black bean sauce. Small soybeans are washed and screened for foreign material, then steamed and fermented in an aerobic environment using the fungus *Aspergillus oryzae*. The resulting fermented black bean (the fermentation process blackens the beans) is washed, dried, screened for any foreign materials and packaged under vacuum until it is used in the extraction stage.

16. Fermentation using the fungus *Aspergillus oryzae* is a well established procedure used in the production of soy sauce, sake and miso. The applicant is of the view that the exact fermentation conditions that lead to a high quality product without risk of toxic by-product formation have been established over years of industrial experience thereby ensuring the safety of the NI.

17. The fermented beans are milled and then suspended in boiling water. The aqueous phase is separated from the insoluble fraction following a series of purification steps before being concentrated and spray dried to create the final product, which is a pale brown powder. The applicant states that the process is typical of the industry and follows conventional extraction procedures.

18. The applicant contends that there are no potential hazards associated with the processes employed in the production of the NI and no hazardous materials are formed under proper, regulated manufacturing conditions using *Aspergillus oryzae*. The procedures involved in the manufacture of the NI follow the principles of HACCP and GMP. The process and intermediate products are monitored routinely to ensure that the NI meets the specification. If the final product does not meet the required specification, it is discarded.

19. The manufacture of the NI complies with principles laid out in Regulation (EC) No 852/2004 on the hygiene of foodstuffs at all stages of the production process. The preparation of the NI in powder form lowers the amount of moisture to <15% thereby reducing the potential for microbial growth and increasing the shelf life of the product.

20. In response to the Committee's request for further information on the similarity of the heat-treated NI to black bean sauce, the applicant provided examples of recipes involving the use of fermented black beans to make a sauce. The applicant notes that although the production of the NI involves 2 separate heat treatments, the second treatment is the sterilisation step and is a controlled treatment performed over a short period of time (135°C, 15 seconds). By comparison, the one heat treatment involved in preparing fermented black bean sauce involves higher temperatures and less controlled conditions. Nevertheless, the applicant was of the view that any general effects of heat treatment on the protein components will be consistent between the sauce and the extract.

21. In response to a further request from the Committee the applicant provided the results of HPLC analysis to demonstrate the effect of fermentation and hot water treatment on the protein content of the product. The chromatograms obtained for the NI and fermented black bean paste were very similar and exhibited peaks consistent with low molecular weight peptides and amino acids. The applicant concluded that these results confirm there are no significant differences in composition between the two products.

22. The Committee also requested further details of the fermentation conditions used in the manufacture of the black beans and clarification as to whether the black bean fermented product is the same as the one used to create traditional black bean sauce. The applicant states that fermentation is carried out in an aerobic environment at 30°C over 7 days using the fungus *Aspergillus oryzae*. The fermentation is stopped by cooling before the mixture is washed with water and dried in a dedicated drying room. The applicant confirms that the fermentation process used in the production of fermented black bean sauce and paste is identical to that used in the manufacture of the NI.

***Discussion**: The Committee was satisfied that the production process for the NI did not give cause for concern, compared with the process for traditional black bean sauce. Members also noted that because the raw material in the fermentation process can only be slowly utilised by the fungus, the culture is unlikely to encounter conditions that could result in secondary metabolites. The Committee was content that the additional HPLC chromatograms provided by the applicant indicated that the peptide and protein compositions of the NI and black bean paste were similar.*

III. History of the organism used as a source of the novel food

Application dossier, pp 18-21

23. The raw material used to prepare fermented black bean is a non-GM variety of soybean (*Glycine max*) which has been widely used in the Sichuan province of China for centuries where it is known as the small yellow bean (the fermentation process blackens the beans). The plant grows annually and is native to East Asia, and as it produces much smaller beans than those usually cultivated it is viewed to be a niche variety and as such has not been subject to genetic modification.

24. The soybeans used by the applicant are cultivated by contract farmers in the Sichuan province. There are no agro-chemicals used on the crop and there are no potential sources of pollution in the vicinity of the contract farms. During harvesting, countermeasures are employed to reduce possible contamination with foreign materials (e.g. stone, chips and husk) and the storage facilities are maintained to appropriate hygiene standards which are fully documented to ensure traceability.

25. The fermentation of the small soybean with *Aspergillus oryzae* has been used for hundreds of years in the production of soy sauce, miso and sake. *Aspergillus oryzae* is a member of the *Aspergillus flavus* group and has undergone extensive selection over the years in order to produce the current strains that are adapted for use in fermentation processes.

26. The fermented black bean product from which the NI is obtained has a long history of safe use in China and more recently in Europe as a component of many Chinese dishes.

***Discussion**: The Committee accepted the applicant's view that small yellow soybean and Aspergillus oryzae have a long history of use throughout the world in the production of products such as soy sauce, miso and sake.*

IX. Anticipated intake/extent of use of the novel food

Application dossier, pp 22-26

27. The applicant intends that their NI will be used during a meal in order to hinder the digestion of carbohydrates in the small intestine.. Any health claims that are attributed to the consumption of the NI are not considered as part of this application. Any health or nutrition claims that may be made for this ingredient would be subject to a separate authorisation procedure under regulation (EC) 1924/2006

28. The NI will be marketed in food supplement-type products at levels that would not exceed 4.5 g per daily serving/dose. The form of the supplement may vary (e.g. tablet or tea/soup-style) but the applicant has stated that all products will be clearly marked with an indication of dose of the NI to indicate that the NI should only be consumed with food. The applicant anticipates that consumers will be familiar with this type of food supplement form and will follow the directions for use. In further information submitted to the Committee, the applicant advised that all powdered tea/soup formulations are specifically designed for consumption by mixing with hot water only and are not intended for addition to other foods.

29. The applicant considers that, as the NI will only be available in clearly marked food supplement products, presentation to the consumer will be controlled to provide a dose of the NI that would not exceed 4.5 g per serving/dose per day.

30. The applicant provides intake data from the UK provided as part of the “Concise European Food Consumption Database” to estimate the worst case scenario whereby adult consumers (16 to 64 years) of teas and soups consume these types of products as part of their conventional food intake. Typical UK food portion sizes for a “mug of tea” of 240 g/serving and “cup-a-soup” type products of 215 g/serving have been used to convert the recorded consumption of these foods to servings/day, which are then translated into NI consumption based on the maximum proposed use level of 4.5 g/serving.

31. The mean estimated total intake of the NI consumed as conventional “cup-a-soup” type product and as a tea (assuming all consumption in this category is in the form of tea) are less than 4.5 g and 13.5 g respectively (equivalent to <1 and approximately 3 servings, respectively). The highest level estimated consumption (97.5th percentile of users) of the NI is 4.5 g and 36 g, respectively (equivalent to 1 or 8 servings approximately).

32. The applicant considers that it is unlikely that consumers will be high level consumers of both “vegetable soups” and “tea, coffee and cocoa” and in this assessment it is assumed that the highest level consumers would be high consumer of one category and only average consumers for the other. For high level consumer of “soups” and average for “tea” and vice versa result in estimated total exposure to the NI of 18 g and 40.5 g, respectively. In addition, the applicant points out that the NI is intended only to be consumed with food, which would eliminate any intake associated with the casual consumption of tea or soup between meals.

33. A NOAEL of 2,500 mg/kg bodyweight/day of NI (see paragraph 49 below) equates to 150 g/day of the NI for a typical 60 kg adult. The average intakes of approximately 3 servings of “tea, coffee and cocoa” and less than half a serving for “vegetable soups” combined lead to a maximum intake of less than 4 servings or less than 18 g/day of the NI and an 8 fold safety factor. The highest estimated consumption of 40.5 g/day gives a 3.7–fold safety factor.

***Discussion**: The Committee considered that the consumption of the NI at the proposed levels of incorporation did not raise any specific safety concerns, based on the safety data provided and the history of consumption of traditional fermented black beans (see below). The Committee sought clarification from the applicant on the meaning of the term ‘nutritional support’ which is used in the company’s documentation and accepted that this term was used only to convey that the anticipated consumption of the NI is during a meal rather than on its own.*

X. Information from previous human exposure to the novel food or its source

Application dossier, pp 27-28

34. Fermented black beans have been widely consumed as a traditional seasoning in China for the last 1000 years and in Europe they have been consumed in Chinese dishes containing “black bean sauce” or "black bean paste” as a seasoning typically at levels of around 15 g per serving.

35. In accordance with the terms of Directive 2003/89/EC all products containing the NI will be clearly labelled as made from soya or soybeans. The applicant is of the view that as the NI will only be used in food supplements, the presence and origin of the ingredient will therefore be clearly apparent to the consumer.

36. The applicant states that proteins in soya bean, particularly the ‘storage proteins’ vicillin and legumin, are thought to be responsible for the allergic response of certain individuals to soya-containing products. During the fermentation process, degradation may occur to form fragments that do not exhibit the same allergenic response in susceptible individuals. This does not alter the requirement to label products containing the NI in accordance with Directive 2003/89/EC, unless the applicant applies for an exemption under that Directive.

37. The NI is currently approved as a Food for Specific Health Use (FOSHU) in Japan where it is sold through its direct mail channel in the same supplement-style products proposed for the EU. In post-marketing monitoring conducted by the manufacturer in 2005, no adverse effects were reported or have been reported to date. In response to a request from the Committee regarding the reporting of adverse effects if the product is marketed in the EU, the applicant has advised that post-market monitoring will be conducted via the manufacturer's helpline to ensure any complaints of adverse effects can be communicated to the applicant and to the ingredient manufacturer.

***Discussion**: The Committee was satisfied that yellow soybean products have been consumed as a traditional seasoning in China for many years and that there were no concerns regarding general safety of the beans. The Committee noted that the applicant will provide a mechanism for EU consumers to report any adverse effects from consuming the NI. The Committee requested that the results of any post-market monitoring should be reported after an interval of 18 months.*

XI. Nutritional information on the novel food

Application dossier, pp 29-32

38. In terms of 'nutritional safety' the applicant considers that the NI is equivalent to fermented black beans and fermented black bean paste which have been on the market in the EU for many years.

39. One portion of black bean sauce in a dish would generally contain 15 g of fermented black beans and on extraction; 15 g of fermented black bean sauce corresponds to 4.5 g of the NI. The applicant provides a comparison of nutrient profiles for 15 g fermented black beans and 4.5 g of NI in Table XI.A-1 of the dossier. The applicant states that, whilst the relative amount of each nutrient changes during processing because the water-insoluble fraction is removed, no chemical modification is involved and hence the nutrients that are present in 4.5 g of the extract are also present in the same amounts in 15 g of the unextracted material.

40. The applicant suggests that the NI, consumed as a food supplement with a meal, can delay carbohydrate digestion in the small intestinal tract. The NI is purported to have the ability to inhibit the activity of the alpha-glucosidase enzyme so limiting the breakdown of carbohydrates and the subsequent formation of glucose and fructose (monosaccharides). The applicant states that undigested carbohydrates or disaccharides are then excreted rather than absorbed by the body therefore potentially offering assistance in weight control regimes.

41. For nutrition labelling purposes, 100g of the NI provides 60 g protein (minimum 55 g), no more than 1 g fat and 25 to 30 g of carbohydrate.

***Discussion**: Members accepted that the nutritional properties of the NI did not give cause for concern. The Committee noted that undigested fibre is likely to be fermented in the gut and referred to a recent review by the Scientific Advisory Committee on Nutrition (SACN)[1] which found limited evidence for the contribution of fibre to weight control.*

XII. Microbiological information on the novel food

Application dossier, pp 33-34

42. The production process for the NI involves sterilisation and filtration of the aqueous extract in order to minimise the risk of microbial contamination. The moisture content of the product in the powder form is <7% and the applicant is of the view that the potential for microbial growth is therefore limited. The manufacturing site has a certified HACCP system in place and all procedures comply with GMP as further assurance of the quality of the NI.

43. The applicant provides the results of an independent microbiological screen on the same three production batches of the NI discussed in Section I (Table XII.A in the dossier). No significant contamination with bacteria, mould, yeast or *E. coli* was detected in any of the batches tested.

***Discussion**: The Committee was of the view that the microbiological safety of the NI had been demonstrated and was reassured that the manufacturing site has a certified HACCP system in place and that all procedures comply with GMP.*

XIII. Toxicological information on the novel food

Application dossier, pp 35-52

44. The NI has undergone a number of toxicological analyses which were conducted by the applicant company. The NI was evaluated for acute oral toxicity in mice in an unpublished study where the LD_{50} was considered to be >5000 mg/kg body weight.

45. In a 28-day subacute/subchronic toxicity study of the NI, 4-week old rats were given doses of 250, 1000, and 2500 mg/kg body weight of the NI. No clinical signs or changes in body weight or food consumption related to the administration of the NI were observed. Although significant decreases in corpuscular haemoglobin and mean corpuscular volume for males in the 1000 mg/kg group were observed, these changes were thought to be unrelated to the test substance because no dose-dependant effects were noted. Although unilateral pelvic dilation was observed in the kidney of one male at the 2500 mg/kg dose group, these changes were considered to be spontaneous. No other significant changes were seen upon

[1] Narrative Synthesis of Health Effects of Potential Dietary Fibre Components - 13th October 2008 (http://www.sacn.gov.uk/pdfs/narrative_synthesis_paper_final.pdf)

histopathological examination. The NOAEL for the NI was considered to be more than 2500 mg/kg in males and females.

46. No studies that addressed the reproductive and developmental toxicity of the NI, or its mutagenic or genotoxic potential, were found in the published scientific literature. However, the applicant has provided a summary of a reverse mutation assay and an *in vivo* micronucleus test which indicated that the NI was neither mutagenic or genotoxic

47. The NI was also evaluated in an in vivo micronucleus test in rats which were administered the NI (10 mL/kg doses) by oral gavage on 2 successive days. Control animals received water at the same volume. A single 0.4 mg/mL dose of mitomycin C (MMC) was administered intraperitoneally (dosing volume 10 mL/kg) as the positive control substance. No dose-dependant increase was observed in the incidence of polychromatic erythrocytes with micronuclei among the NI-treated groups, whereas a significant increase in the incidence of polychromatic erythrocytes with micronuclei (4.92%) was observed in the positive control group treated with MMC.

48. The activity of the NI was evaluated in tissue, animal, and anti-infective in vitro assays as part of a pharmacological screen (PharmaScreen, MDS Panlabs Pharmocology Service, USA). The NI did not produce automatic signs or effects on the central nervous system, cardiovascular system, and gastrointestinal system. No metabolic effects were seen, nor were any indications of allergy or inflammation observed. No significant activity was observed at dose levels and concentrations tested. No microbiological pathogens were detected. A limited number of pharmacological studies in laboratory animals were also identified. No adverse effects were reported in rats and mice associated with the administration of the NI. The applicant provides a summary of studies examining the pharmacological effects of the NI in Table XIII.A. 6-1.

49. Human studies examining the effect of the NI on carbohydrate digestion following a meal in healthy, hyperlipidemic and diabetic subjects have also been summarised as part of this dossier. Patients were monitored for changes in various haemological and biochemical parameters, body weights and subjective side effects. The applicant is of the view that the absence of major adverse effects offers additional support for the safety of the NI. A summary of the clinical trials with the NI is provided in Table XIII.B.1-1. The applicant notes that no gastrointestinal effects were seen in clinical studies with the NI. The applicant also states that although soybeans can inhibit gastrointestinal proteases and therefore induce diarrhoea or abdominal pain, the NI did not demonstrate protease inhibition or cause this effect.

***Discussion**: The Committee was content that the toxicological assessment carried out by the applicant on the NI showed no evidence of adverse effects*

***Overall Discussion**: The Committee accepted that fermented black bean products are widely available on the market and have been consumed for many years in the form of soy sauce, miso and sake. Because the NI is obtained from fermented black bean using the same production process used for the production of black bean sauce, the Committee was content that no adverse effects are expected from consuming the NI. In addition the Committee was reassured that the fermentation conditions employed would be unlikely to give rise to the production of secondary metabolites.*

CONCLUSION

50. The Advisory Committee on Novel Foods and Processes is satisfied by the evidence provided by CBC Co Ltd that the range of uses for its Touchi extract is acceptable, subject to the applicant's adherence to the proposed specification and the production parameters described above. The Committee also wishes to note that foods containing this novel ingredient should be labelled in accordance with existing legislation and should not make claims that are likely to mislead consumers.

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