Risk assessment of "other substances" – D-Ribose

Opinion of the Panel Food Additives, Flavourings, Processing Aids, Materials in Contact with Food and Cosmetics of the Norwegian Scientific Committee for Food Safety

Risk assessment of "other substances" – D-ribose

Authors preparing the draft opinion

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Assessed and approved

The opinion has been assessed and approved by Panel on Food Additives, Flavourings, Processing Aids, Materials in Contact with Food and Cosmetics. Members of the panel are: Inger-Lise Steffensen (Chair), Ellen Bruzell, Berit Granum, Ragna Bogen Hetland, Trine Husøy, Jens Rohloff, Trude Wicklund.

(Panel members in alphabetical order after chair of the panel)

Acknowledgment

Competence of VKM experts

Persons working for VKM, either as appointed members of the Committee or as external experts, do this by virtue of their scientific expertise, not as representatives for their employers or third party interests. The Civil Services Act instructions on legal competence apply for all work prepared by VKM.

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Summary

The Norwegian Scientific Committee for Food Safety (Vitenskapskomiteen for mattrygghet, VKM) has, at the request of the Norwegian Food Safety Authority (Mattilsynet; NFSA), assessed the risk of "other substances" in food supplements and energy drinks sold in Norway. VKM has assessed the risk of doses in food supplements and concentrations in energy drinks given by NFSA. These risk assessments will provide NFSA with the scientific basis while regulating the addition of "other substances" to food supplements and other foods.

"Other substances" are described in the food supplement directive 2002/46/EC *as substances other than vitamins or minerals that have a nutritional and/or physiological effect*. It is added mainly to food supplements, but also to energy drinks and other foods. VKM has not in this series of risk assessments of "other substances" evaluated any claimed beneficial effects from these substances, only possible adverse effects.

The present report is a risk assessment of D-ribose, and it is based on previous risk assessments and articles retrieved from a literature search.

According to information from NFSA, D-ribose is an ingredient in food supplements sold in Norway. NFSA has requested a risk assessment of 3100 and 6200 mg/day of D-ribose in food supplements for the age groups children (10 to <14 years), adolescents (14 to <18 years) and adults (>18 years).

Other sources of D-ribose, such as foods and cosmetics, have not been included in the present risk assessment.

D-ribose is a component of the genetic material RNA and is synthesized in all living cells via the pentose phosphate pathway. D-ribose is also a structural component of adenosine triphosphate (ATP), the primary source of cellular energy and a key component of riboflavin (e.g. vitamin B_2). The estimated endogen synthesis of D-ribose is referred to be from 2.7 g per day (women) to 16.5 g per day (men). D-ribose is available in small amounts in the diet via ripe fruits and vegetables. It is also an ingredient in food supplements, some so-called energy drinks and in cosmetics as skin conditioner and humectant.

Orally administered D-ribose is absorbed in the small intestine by passive diffusion. Absorption rates after oral ingestion of doses up to 200 mg/kg bw per hour (administered for 5 hours) have been shown to range from 87.8 to 99.8% in humans.

No serious adverse health effects were identified at doses up to 20 g per day as reported in the human studies included in this opinion.

Based on a subchronic oral toxicity study in rats, no observed adverse effect levels (NOAELs) of 3.6 and 4.4 g/kg bw per day in males and females were derived. The NOAELs were based on a statistically significant decrease in body weight. In another study in rats, the NOAELs

for embryo toxicity/teratogenicity of D-ribose were 3.6 and 4.6 g/kg bw per day based on individual females. This NOAELs were primarily based on a statistically significantly higher incidence of one or multiple wavy ribs in the mid- and high-dose groups compared to control animals.

No studies on children (10 to <14 years) and adolescents (14 to <18 years) were identified. Based on the included literature there was no evidence indicating that age affects tolerance for D-ribose. Therefore, in this risk characterisation a tolerance as for adults, based on body weight, were assumed for these age groups.

The values used for comparison with the estimated exposure in the risk characterization are 20 g per day (corresponding to 286 mg/kg bw per day in a 70 kg adult) considered to be without appreciable health risk for most healthy adults and the NOAEL of 3.6 g/kg bw per day from the subchronic toxicity and embryotoxicity/teratogenicity studies in rats.

From a daily dose of 3100 mg or 6200 mg of D-ribose, the intake levels are 71.4, 50.6 and 44.3 mg/kg bw per day and 142.6, 101.1 and 88.6 mg/kg bw per day for for children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years), respectively.

The calculated MOE values from the rat study for a daily intake of 3100 mg per day were 50.4, 71.1 and 81.3 for children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years), respectively. The calculated MOE values for a daily intake of 6200 mg per day were 25.2, 35.6 and 40.6 for children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years), respectively. In this case, MOE values below 100 are regarded as acceptable since D-ribose is present in all cells in the body and the daily doses from food supplements are in the same order as the endogenous production, which ranges from 2.7 g per day (women) to 16.5 g per day (men) (Bioenergy Life Science Inc., 2008).

VKM concludes that it is unlikely that daily doses of 3100 mg or 6200 mg D-ribose in food supplements causes adverse effects in children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years).

Short summary

The Norwegian Scientific Committee for Food Safety (VKM) has, at the request of the Norwegian Food Safety Authority, assessed the risk of intake of 3100 mg pe day and 6200 mg per day of D-ribose in food supplements. In the risk characterization, the values used for comparison with the estimated exposure are 20 g per day (corresponding 286 mg/kg bw per day in a 70 kg adult) from the human studies and a NOAEL of 3.6 g/kg bw per day from the subchronic toxicity and embryotoxicity/teratogenicity studies in rats.

VKM concludes that it is unlikely that daily doses of 3100 mg or 6200 mg D-ribose in food supplements causes adverse effects in children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years).

Sammendrag på norsk

På oppdrag for Mattilsynet har Vitenskapskomiteen for mattrygghet (VKM) vurdert risiko ved tilsetting av «andre stoffer» i kosttilskudd og energidrikk som selges i Norge. VKM har risikovurdert ulike doser brukt av kosttilskudd og konsentrasjoner i energidrikker oppgitt fra Mattilsynet. Disse risikovurderingene vil gi Mattilsynet vitenskapelig grunnlag for å regulere andre stoffer.

«Andre stoffer» er beskrevet i kosttilskuddsdirektivet 2002/46/EC som *stoffer som har en ernæringsmessig og/eller fysiologisk effekt, og som ikke er vitaminer og mineraler*. De tilsettes i hovedsak til kosttilskudd, men også til energidrikker og andre næringsmidler. I disse risikovurderingene har VKM ikke sett på påståtte gunstige helseeffekter, men kun vurdert mulige negative helseeffekter.

Denne rapporten er en risikovurdering av D-ribose, og den er basert på tidligere risikovurderinger og artikler hentet fra et litteratursøk.

I følge informasjon fra Mattilsynet er D-ribose en ingrediens i kosttilskudd og energidrikker som selges i Norge. Oppdraget fra Mattilsynet var å risikovurdere inntak på 3100 mg per dag og 6200 mg per dag av D-ribose i kosttilskudd.

Andre kilder til D-ribose, som mat og kosmetikk, er ikke inkludert i denne risikovurderingen.

D-ribose er en komponent i det genetiske materialet RNA og syntetiseres i alle levende celler via pentose-fosfat reaksjonsveien. D-ribose er også en strukturell komponent av adenosin trifosfat (ATP), den primære energikilden i alle celler, og en viktig komponent i riboflavin (f.eks. vitamin B$_2$). Estimert endogen syntese av D-ribose er beskrevet å være fra 2,7 g/dag (kvinner) til 16,5 g per dag (menn). D-ribose finnes i små mengder i moden frukt og grønnsaker. Det er også en ingrediens i kosttilskudd, enkelte såkalte energidrikker og i kosmetikk som hudkremer og fuktighetsbevarende produkter.

Oralt administrert D-ribose blir absorbert i tynntarmen ved passiv diffusjon. Absorpsjonsgrad etter oralt inntak av doser på opptil 200 mg/kg kroppsvekt per time (administrert i 5 timer), er i området 87,8 til 99,8 % hos mennesker.

Ingen alvorlige helseeffekter ble identifisert ved doser opp til 20 g per dag i de humane studiene som er inkludert i denne vurderingen.

Basert på en subkronisk oral toksisitetsstudie i rotter ble det funnet en NOAEL-verdi på 3,6 g/kg kroppsvekt per dag for hanner og 4,4 g/kg kroppsvekt per dag for hunner. NOAEL-verdiene ble satt på bakgrunn av en statistisk signifikant nedgang i kroppsvekt i de to høyeste dosegruppene. I en annen studie i rotter ble det funnet NOAEL-verdier for embryotoksisitet/teratogenisitet på 3.6 og 4.6 g/kg kroppsvekt per dag i individuelle hunner basert på en statistisk signifikant høyere forekomst av en eller flere bølgeformede ribben.

I denne risikovurderingen har VKM brukt en dose på 20 g per dag (tilsvarende 286 mg/kg kroppsvekt per dag for en voksen person på 70 kg), som anses for å være uten vesentlig helserisiko for friske voksne, og en NOAEL på 3,6 g/kg kroppsvekt per dag fra en subkronisk toksisitetsstudie og en studie på embryotoksisitet/teratogenisitet i rotter.

En daglig dose på 3100 mg eller 6200 mg av D-ribose tilsvarer 71,4, 50,6 og 44,3 mg/kg kroppsvekt per dag eller 142,6, 101,1 og 88,6 mg/kg kroppsvekt per dag for henholdsvis barn (10 til <14 år), ungdom (14 til <18 år) og voksne (≥18 år).

Beregnet ratio mellom en NOAEL-verdi på 3,6 g/kg kroppsvekt per dag, og inntak av D-ribose fra kosttilskudd (MOE-verdier) ved et daglig inntak på 3100 mg var 50,4, 71,1 og 81,3 for henholdsvis barn (10 til <14 år), ungdom (14 til <18 år) og voksne (≥18 år). For et daglig inntak på 6200 mg var MOE-verdiene 25,2, 35,6, og 40,6 for henholdsvis barn (10 til <14 år), ungdom (14 til <18 år) og voksne (≥18 år). I dette tilfellet ansees MOE-verdier under 100 som akseptable siden D-ribose er tilstede i alle kroppens celler og de daglige dosene fra kosttilskudd er i samme størrelsesorden som den endogene produksjonen, som er oppgitt å være 2,7 g per dag i kvinner og 16,5 g per dag i menn.

VKM konkluderer at det er usannsynlig at en daglig dose på 3100 mg eller 6200 mg av D-ribose fra kosttilskudd forårsaker negative helseeffekter hos barn (10 til <14 år), ungdom (14 til <18 år) og voksne (≥ 18 år).

Kort sammendrag

På oppdrag fra Mattilsynet har Vitenskapskomiteen for mattrygghet (VKM) vurdert helserisiko ved inntak av 3100 mg per dag og 6200 mg per dag av D-ribose fra kosttilskudd. I risikokarakteriseringen ble eksponeringen for D-ribose fra kosttilskudd sammenlignet med et inntak på 20 g per dag (tilsvarer 286 mg/kg kroppsvekt per dag i en 70 kg voksen) fra studier i mennesker og en NOAEL på 3,6 g/kg kroppsvekt per dag fra subkroniske studier i rotter med endepunktene generell toksisitet og embryotoksisitet/teratogenisitet.

VKM konkluderer at det er usannsynlig at en daglig dose på 3100 mg eller 6200 mg av D-ribose fra kosttilskudd forårsaker negative helseeffekter hos barn (10 til <14 år), ungdom (14 til <18 år) og voksne (≥ 18 år).

Abbreviations and glossary

Abbreviations

ACNFP	- Advisory Committee on Novel Foods and Processes, Food Standards Agency, UK
ADME	- Absorption, distribution and excretion
AGE	- Advanced glycation end products
ALP	- alkaline phosphatase
ALT	- alanine aminotransferase
AST	- aspartate aminotransferase
ATP	- adenosine triphosphate
bw	- body weight
CBC	- complete blood count
EFSA	- European Food Safety Authority
FDA	- Food and Drug Administration (US)
GGT	- gamma glutamyltransferase
GPT	- glycated plasma proteins
GRAS	- Generally Recognised as Safe
GSH	- glutathion
Hct	- hematocrit
Hgb	- hemoglobin
MDA	- malondialdehyde
MOE	- Margin of exposure
NFSA	- Norwegian Food Safety Authority [*Norw.*: Mattilsynet]
NOAEL	- no observed adverse effect level
NOEL	- no observed effect level
Plts	- platelet counts
RNA	- ribonucleic acid
VKM	- Norwegian Scientific Committee for Food Safety [Norw.: Vitenskapskomiteen for Mattrygghet]
WBC	- white blood count

Glossary

"Other substances": a substance other than a vitamin or mineral that has a nutritional or physiological effect (The European Parliament and the Council of the European Union, 2006).

"Negative health effect" and "adverse health effect" are broad terms. VKM uses the definition endorsed by EFSA for "adverse effect": a change in morphology, physiology, growth, development, reproduction or life span of an organism, system or (sub)population that results in an impairment of functional capacity, an impairment of the capacity to compensate

for additional stress, or an increase in susceptibility to other influences (EFSA, 2006; WHO, 1994).

Background as provided by the Norwegian Food Safety Authority

«Other substances» are substances other than vitamins and minerals, with a nutritional and/or physiological effect on the body. "Other substances" are mainly added to food supplements, but these may also be added to other foods and beverages, such as sports products and energy drinks. Ingestion of these substances in high amounts presents a potential risk for consumers.

In Norway, a former practice of classification of medicines constituted an effective barrier against the sale of potentially harmful "other substances". Ever since this practice was changed in 2009, it has become challenging to regulate and supervise foods with added "other substances". Meanwhile, in the recent years, the Norwegian market has witnessed a marked growth in the sales of products containing "other substances". In 2011, food supplements containing "other substances" constituted more than 50% of the market share.

While within the European Economic Area, these substances fall under the scope of the European Regulation (EC) No. 1925/2006 on the addition of vitamins, minerals and certain other substances to foods and the European Regulation (EC) No 258/97 concerning novel foods and novel food ingredients, "other substances" remain largely unregulated. In order to ensure safe use of "other substances" many countries have regulated their use at a national level. For example, Denmark regulates these substances in a positive list, i.e. a list of substances with maximal daily doses, permitted for use in food supplements and other foods (FVM, 2014).

The Norwegian Food Safety Authority (NFSA) is working on the establishment of a regulation on the addition of "other substances" to foods at a national level. The regulation will include a list of substances with permitted maximal doses, based on the substances and doses found in products on the Norwegian market. In preparation for a regulation, NFSA has therefore requested the Norwegian Scientific Committee for Food Safety (VKM) to assess the safety of "other substances" found on the Norwegian market. NFSA, in consultation with the industry, has compiled a list of "other substances" found in products marketed in Norway. Only substances with a purity of minimum 50% or concentrated 40 times or more have been included in the list. Substances regulated by other legislations like those for novel foods, food additives, flavourings, foods for special medical purposes etc., have been excluded from the list.

Terms of reference as provided by the Norwegian Food Safety Authority

The Norwegian Food Safety Authority (NFSA) has requested the Norwegian Scientific Committee for Food Safety (VKM) to assess the safety of D-ribose in food supplements at the following doses: 3100 mg/day and 6200 mg/day.

NFSA requested VKM to assess the safety of "other substances" (in accordance to the guidance document developed in Phase 2) at the doses specified (Phase 3). The safety assessments of "other substances" present in food supplements shall be carried out for a general population, ages 10 years and above.

Assessment

1 Introduction

"Other substances" are described in the food supplement directive 2002/46/EC *as substances other than vitamins or minerals that have a nutritional and/or physiological effect*, and may be added to food supplements or e.g. energy drinks (The European Parliament and the Council of the European Union, 2006).

This risk assessment regards the substance D-ribose *per se* and no specific products.

VKM has in this series of risk assessments of "other substances" not evaluated documentation of any claimed beneficial effects from these substances, but merely possible adverse effects at specified doses used in Norway. Thus, potential high intake consumer groups of the substance may not be identified and included in this assessment.

According to information from the Norwegian Food Safety Authority (NFSA), D-ribose is an ingredient in food supplements purchased in Norway. NFSA has requested a risk assessment of the intake of 3100 mg and 6200 mg D-ribose per day from food supplements. The total exposure to D-ribose from other sources than food supplements, such as foods and cosmetics, is not included in the risk assessment.

D-ribose is available in small amounts in the diet via ripe fruits and vegetables (Dhanoa and Housner, 2007; Griffiths et al., 2007a; Griffiths et al., 2007b). It is a five carbon monosaccharide present in all living cells as it is a component of the genetic material RNA and also involved in cellular metabolism. D-ribose is synthesized in all cells, and it has been estimated that the daily endogenous production of D-ribose ranges from 2.7 g/day (women) to 16.5 g/day (men) (Bioenergy Life Science Inc., 2008). The total dietary intake of D-ribose is not known. In this risk assessment, daily intake of 3100 and 6200 mg/day is assessed.

2 Hazard identification and characterisation

2.1 Literature

The present risk assessment is based on previous risk assessments of D-ribose and articles retrieved from a literature search.

2.1.1 Previous risk assessments

Application for the approval of D-ribose for use as an ingredient in foods, food supplement and food for particular nutritional uses. Food Standard Agency, UK (Bioenergy Life Science Inc., 2008)

The applicant concluded that the available evidence presented in the dossier supports the safety of D-ribose. On the basis of the toxicological data available, it was concluded that D-ribose does not present a significant health risk to any age group in the population at the proposed intake levels described in the dossier (highest mean and 97.5th percentile intakes were observed in children, estimated to be 3.8 and 11.0 g/person per day, respectively). The toxicological evaluation of safety of D-ribose was based on metabolic data in animals and humans, results from a subchronic oral toxicity study and a reproductive/teratology study in rats, the results of genotoxicity studies and results from human studies.

Draft initial opinion on an application under the novel foods regulation for D-ribose. Food Standards Agency, UK (Advisory committee on novel foods and processes, 2016)

The Advisory Committee on Novel Foods and Processes (ACNFP), an independent committee appointed by the Food Standards Agency, UK, has evaluated an application from Bioenergy Life Science Inc. regarding authorization of D-ribose produced by the bacterium *Bacillus subtilis* as a novel ingredient in EU. The applicant intends to use their D-ribose product in a variety of foods including food supplements. The Committee was content with the intake data presented by the applicant and the use of a NOAEL value of 4 g/kg bw derived from a 13-week study in rats for the risk assessment of the intended use of D-ribose. Furthermore, the Committee was satisfied that D-ribose is only proposed for addition to foods that contain other carbohydrate energy sources, and that the content of D-ribose in reduced calorie drinks would only be 1 g per serving. The Committee recommended labelling food supplements containing D-ribose not to be taken on an empty stomach as a warning against possible hypoglycaemic effects. In their draft initial opinion, the ACNFP concluded that the Committee was satisfied by the evidence provided by Bioenergy Life Science Inc. that D-

ribose was acceptable as a novel food ingredient, subject to the applicant's adherence to the proposed labelling requirements.

The Committee has asked for comments on the draft opinion, deadline was 1 February 2016 (ACNFP, 2016).

Substances Generally Recognised as Safe. USA (GRAS, 2008)

The United States Food and Drug Administration (U.S. FDA) concluded that D-ribose is classified as GRAS for use as an ingredient in specified foods, given the intended conditions of use and provided that D-ribose is used in conjunction with an additional carbohydrate energy source. Low-calorie beverages are excluded from the intended uses (U.S. FDA, 2008).

2.1.2 Literature search

Literature searches were performed in Embase and Medline in order to retrieve publications on adverse effects caused by D-ribose. These databases were chosen to ensure comprehensive study retrieval. The literature searches were performed in January 2016. The search strategy is included in Appendix 1.

2.1.2.1 Publication selection and data extraction

The literature search identified 48 articles. In the primary screening, titles and abstracts of all publications retrieved were independently screened against the inclusion criteria checklist.

Inclusion criteria checklist:
- Adverse effects in relation to the substance alone are addressed
- Route of exposure for humans is oral
- Route of exposure for animals is oral, in addition, subcutaneous exposure is included if the toxicokinetic is equal to oral exposure
- Human studies are performed in apparently healthy individuals or patient groups assumed to have normal absorption and metabolism of the assessed substance
- Animal model studies address adverse effects relevant to human health

The inclusion criteria checklist was developed by members of the Panel on Food Additives, Flavourings, Processing Aids, Materials in Contact with Food and Cosmetics and the Panel on Nutrition, Dietetic Products, Novel Food and Allergy. Articles that did not appear to meet the inclusion criteria were excluded from further analysis. In situations where it was unclear whether the publication was of relevance to the study, it was retained for further screening. The primary screening was performed independently by two persons.

The full text of articles that passed the primary screening was retrieved for secondary screening. In this screening, the full text articles were reviewed and compared against the inclusion criteria checklist. The secondary screening was performed by one person.

The secondary screening resulted in 5 full text articles. Additionally, 4 studies from manual search were identified and included. A final total of 9 publications were identified and included in the results in this report (see Figure 2.1.2.1-1).

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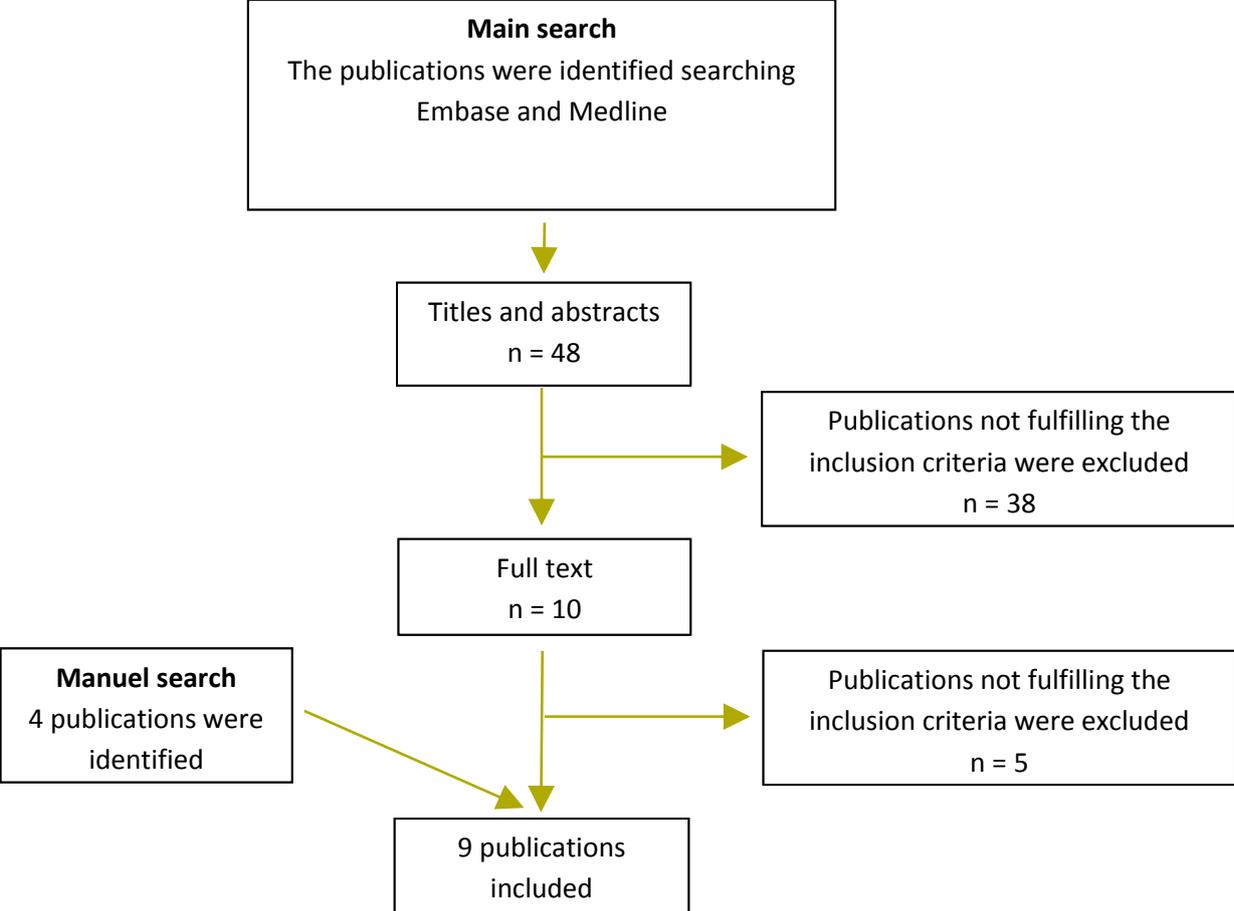
Figure 2.1.2.1-1   Flowchart for the literature search for D-ribose and the subsequent publication selection.

2.2 General information

2.2.1 Chemistry

D-ribose (CAS no. 50-69-1, EINECS no. 200-059-4) is a naturally occurring carbohydrate, a five carbon monosaccharide. It is an aldopentose due to the aldehyde functional group, and the molecular formula is $C_5H_{10}O_5$. The structural formulas are shown in Figure 2.2.1-1.
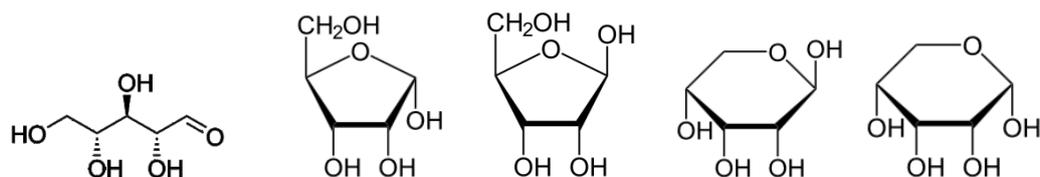

Figure 2.2.1-1 The structural formulas of the 5 isomers of D-ribose (from left: open chain, α-D-ribopyranose, β-D-ribopyranose, α-ribofuranose and β-ribofuranose).

2.2.2 Occurrence

D-ribose is a component of the genetic material RNA and is synthesized in all living cells via the pentose phosphate pathway. D-ribose, in the form of ribonucleoside diphosphate, is converted to deoxyribonucleoside diphosphate, precursor molecules for DNA. D-ribose is also a structural component of adenosine triphosphate (ATP), the primary source of cellular energy and a key component of riboflavin (i.e. vitamin B_2) (Griffiths et al., 2007a; Griffiths et al., 2007b). The estimated endogen synthesis of D-ribose is 2.7 g/day (women) and 16.5 g/day (men) (Application for the approval of D-ribose, 2008). D-ribose is available in small amounts in the diet via ripe fruits and vegetables (Dhanoa and Housner, 2007). It is also an ingredient in food supplements, some so-called energy drinks and in cosmetics as skin conditioner and humectant.

2.3 Absorption, distribution, metabolism and excretion (ADME)

Orally administered D-ribose is generally thought to be absorbed in the small intestine by passive diffusion. Absorption rates after oral ingestion of doses up to 200 mg/kg bw per hour (administered for 5 hours) have been shown to range from 87.8 to 99.8% in humans (Gross et al., 1989). Diffusion capacity of D-ribose in the small intestine can be exceeded both in humans and rodents, depending on the amount ingested and other dietary components. Unabsorbed D-ribose passes on to the large intestine where it undergoes fermentation and/or excretion in faeces (Griffiths et al., 2007b). Once absorbed, D-ribose is phosphorylated and enters the pentose phosphate pathway of glucose metabolism as a substrate for purine and pyrimidine biosynthesis. When pentose phosphates are not needed for purine nucleotide synthesis in muscle, or when ribose is present in excess amounts,

pentose phosphates are recycled through glycolysis mainly in the liver via conversion into fructose-6-phosphate, fructose-1,6-bisphosphate (to a lesser degree), and glyceraldehyde-3-phosphate, eventually to form CO_2 and water and yielding energy via ATP turnover. The metabolism of D-ribose in humans following intravenous administration indicates that D-ribose is rapidly and extensively metabolised via the pentose phosphate pathway. These data also suggests that D-ribose induces a lowering of blood glucose, presumably by the competitive effect on phosphoglucomutase, thereby preventing metabolism of glycogen to glucose in the liver (Segal and Foley, 1958).

2.4 Toxicological data/Adverse effects

2.4.1 Human studies

An overview of the included human studies of adverse health effects of D-ribose is given in Table 2.4.1-1.

Table 2.4.1.1 An overview of human studies investigating D-ribose and adverse health effects. (M=male; F=female).

Reference	Study design/participant characteristics	Country	Number in treatment group		Dose	Main endpoint	Study duration	Adverse effect
			D-ribose	**Control/placebo**				
Seifert et al. (2009)	Double-blind, placebo-controlled crossover (experienced recreational cyclists)	USA	7 (gender not referred)	7 (gender not referred)	2 x 7 g D-ribose on training day, in total 14 g/day. Placebo: flavoured water	Role of D-ribose on oxidative stress during hypoxic exercise (hemoglobin, uric acid, glucose and glutathione (GSH) in blood, creatinine and malondialdehyde (MDA) in urine)	1 day training + at least 1 week wash out + 1 day training (total ≥ 9 days)	All subjects tolerated the test substance without adverse effects
Seifert et al. (2008)	Intervention study without control group	USA	19 (12 M and 7 F)	No controls	2 x 10 g D-ribose per day (at breakfast and dinner), in total 20 g/day	Assessment of hematological and biochemical parameters with extended D-ribose ingestion. The following parameters were analysed: complete blood count (CBC), hemoglobin (Hgb), hematocrit (Hct), white blood count (WBC), platelet counts (Plts), albumin, alkaline phosphatase (ALP), gamma glutamyltransferase (GGT), alanine aminotransferase (ALT), aspartate aminotransferase (AST), uric acid, glucose	14 days	There were no adverse physical symptoms or statistically significant differences in hematological or biochemical parameters.
Dunne et al. (2006)	Double-blind, randomized, placebo-controlled (members of auniversity rowing team)	USA	13 (F)	18 (F)	2 x 10 g D-ribose per day (before and after workout), in total 20 g/day for 8 weeks. Placebo: dextrose	Ribose versus dextrose, association with rowing performance	4-days organized and 1 – 2 days self-directed practice per week for 8 weeks	No serious adverse effects were reported. Mild stomach discomfort was more common in the dextrose group
Kreider et al. (2003)	Double-blind, randomized, placebo-controlled (healthy, trained males)	USA	9 or 10 (M) (19 of a total of 20 completed the study)	9 or 10 (M) (19 of a total of 20 completed the study)	2 x 5 g D-ribose per day (morning and evening), in total 10 g/day. Placebo: dextrose	Effects of oral D-ribose supplementation on anaerobic capacity and selected metabolic markers (ammonia, lactate, glucose and uric acid)	5 days	No reports of medical problems/symptoms in post-study questionnaires

Reference	Study design/participant characteristics	Country	Number in treatment group		Dose	Main endpoint	Study duration	Adverse effect
			D-ribose	**Control/placebo**				
't Eijnde et al. (2001)	Double-blind, randomized, placebo-controlled (healthy male physical education students)	Belgium	10 (M)	10 (M)	4 x 4 g D-ribose per day, in total 16 g/day Placebo: maltodextrin	Effect of D-ribose on restoration of muscle ATP after exercise and muscle force and power during training	1 week treatment + 6 weeks wash-out + 1 week treatment (in total 8 weeks)	No negative effects related to ribose supplementation were reported over the duration of the study

The effects of D-ribose on oxidative stress during hypoxic exercise were tested in a double-blinded, cross-over study by studying two markers of free radical production, malondialdehyde (MDA) and glutathione (GSH) (Seifert et al., 2009). Seven experienced recreational cyclists cycled at their lactate threshold for 25 minutes while inhaling 16% O_2 with a subsequent 60 minutes resting period at room air. Subjects ingested either 250 ml flavoured water (placebo) or 7 g of D-ribose in 250 ml of water before and after the exercise session (total of 14 g D-ribose per day). Urinary MDA levels and plasma GSH levels increased significantly during placebo ingestion ($P < 0.05$). After ribose ingestion, MDA levels were maintained at baseline levels whereas GSH levels showed a lesser elevation and subsequent decline. Uric acid and lactate were elevated in both groups. No statistically significant differences in glucose levels were measured in either group. All subjects tolerated both test substances and exercise conditions without adverse effects. The authors concluded that D-ribose demonstrated a beneficial trend in lower MDA and reduced GSH levels under hypoxic stress.

In a study aiming to assess the toxicity of extended consumption of D-ribose in healthy adults, nineteen subjects ingested 20 g D-ribose per day (10 g at breakfast and dinner each day) for 14 days (Seifert et al., 2008). There was no control group. Biochemical and hematological parameters were measured at days 0, 7 and 14. There were no adverse physical symptoms in the subjects completing the study, and no statistical significant changes in blood sample measurements were demonstrated. Measured parameters included complete blood count (CBC), hemoglobin (Hgb), hematocrit (Hct), white blood count (WBC), platelet counts (Plts), albumin, alkaline phosphatase (ALP), gamma glutamyltransferase (GGT), alanine aminotransferase (ALT), aspartate aminotransferase (AST), uric acid and glucose levels. However, both genders demonstrated a mild, but not statistically significant, decrease in serum glucose levels (approx. 0.6 mmol/L and 0.2 mmol/L in females and males, respectively, estimated from Figure 1 in Seifert et al. (2008) over the study. A slight, but not statistically significant, increase in uric acid from day 0 to day 7 (approx. 20 µM/L in both sexes, estimated from Figure 2 in Seifert et al. (2008) was also observed. In summary, the study demonstrated that 20 g per day of D-ribose did not elicit significant adverse hematological or biochemical abnormalities VKM support this conclusion since the observed decreases in serum glucose levels and increases in uric acid were within the normal physiological fluctuations in humans.

The effects of D-ribose versus dextrose supplementation on rowing performance were studied in women collegiate rowers in a double-blind, randomized trial (Dunne et al., 2006). The study lasted for 8 weeks, and the training regimen was based on 4 days organized practice and 1 – 2 days of self-directed practice per week. The supplemental drinks contained 10 g of either D-ribose or dextrose (placebo) in 237 ml of water. Each participant drank 1 drink before and after work-out each day, resulting in a daily dose of 20 g of either D-ribose or dextrose for 8 weeks. Median changes in 2000-meter distance rowing time from baseline to each follow-up time trial (2, 5 and 7 weeks) and end of study at 8 weeks were used as measure for improvement. At 8 weeks, subjects in the dextrose group showed a median improvement of 15.2 sec, whereas the D-ribose group showed a median

improvement of 5.2 sec. Post-study surveillance surveys showed that mild stomach discomfort was more common in the dextrose group than the ribose group (7/14 vs. 1/11, P = 0.041). The groups did not differ statistically significantly in prevalence of any other negative effects (bad taste, diarrhea, nausea, headache, lightheadedness, palpitations or dizziness) or in post-trial exhaustion and recovery.

The effects of oral D-ribose supplementation on anaerobic capacity and selected metabolic markers were studied in a double-blind, randomized, placebo-controlled study in healthy, trained male athletes (Kreider et al., 2003). Anaerobe capacity tests involved 5 min. warming up and two 30 sec. sprint tests separated by 3 min. rest recovery. Tests were performed on a computerized CardiO$_2$™ ergometer cycle and measured on day 0 and day 6. Blood samples were collected immediately after the first and second sprints as well as after 5 min. of recovery of the second sprint. Based on pre-supplementation testing, the subjects were matched according to body mass and anaerobe capacity and assigned to supplement their normal diet with capsules containing 5 g of D-ribose or dextrose (placebo) twice daily for 5 days. Results indicated that oral D-ribose supplementation at this level did not affect anaerobic capacity (peak power, average power, torque and fatigue index) or metabolic markers (lactate, ammonia, glucose and uric acid). Subjects tolerated the supplementation protocol well, with no reports of medical problems/symptoms in post-study questionnaires.

A double-blind randomized study was performed in healthy male physical education students to evaluate the effects of oral ribose supplementation on repeated maximal exercise and ATP recovery after intermittent maximal muscle contractions ('t Eijnde et al., 2001). Muscle power output was measured during dynamic knee extension before (pretest) and after (posttest) a 6-day training period in conjunction with D-ribose (4 doses per day at 4 g/dose) or maltodextrin (placebo) intake. The exercise protocol consisted of two bouts (A and B, separated by a 60 min rest period), and the same exercise protocol was performed twice a day with 3 – 5 hours rest between exercise sessions. After a 6-week washout period, biopsy samples were taken from the vastus lateralis before, immediately after and 24 hours after an exercise bout similar to the pretest. Blood and plasma metabolites (lactate, ammonia, glucose, uric acid and creatine kinase) taken during and after intermittent muscle contractions were similar between treatment groups. The authors concluded that oral ribose supplementation at 16 g per day (4 x 4 g) did not beneficially impact on post-exercise muscle ATP recovery and maximal exercise performance. No side effects related to the D-ribose supplementation were reported over the duration of the study.

2.4.1.1 Interactions

There was no information concerning interactions in the literature reviewed in the present risk assessment. The absence of information in the selected literature does not document an absence of interactions.

2.4.1.2 Allergic sensitisation (including adjuvant effects)

There was no information concerning allergic sensitisation or allergy adjuvant effects in the literature reviewed in the present risk assessment. The absence of information in the selected literature does not document an absence of allergic sensitisation or allergy adjuvant effects.

2.4.2 Animal studies

An overview of the included studies of adverse effects of D-ribose in animals is given in Table 2.4.2-1.

Table 2.4.2-1 An overview of animal studies of adverse effects of D-ribose. (M=male; F=female).

Reference	Study	Dose and number in treatment group		Conclusion with regard to adverse effects
		D-ribose	Control	
Sinatra and Caiazzo (2015)	Effect of D-ribose supplementation on glycated plasma proteins (GPT) in horses Study duration: 17 weeks	Group 1: 30 g/day administered in food once a day (n = 5) (55 mg/kg bw per day) Group 2: 50 g/day administered in food once a day (n=5) (92 mg/kg bw per day)	Baseline level of GPT for each group served as control level	No adverse effects of D-ribose supplementation were observed for the duration of the study. No statistically significant changes in concentrations of GPT compared to baseline were observed during the supplementation period of 17 weeks
Griffiths et al. (2007b)	Oral toxicity study of D-ribose in Wistar rats (OECD Test guideline 408, 1998) Study duration: 13 weeks	5%, 10% and 20% D-ribose in the diet (3.6, 7.6 and 15.0 g/kg bw per day in males, 4.4, 8.5 and 15.7 g/kg bw per day in females) (n = 20 M + 20 F)	Control diet with 0% supplemental D-ribose (n = 20 M + 20 F)	Mean body weight was stastitically significantly decreased in all treated animals relative to control from day 7 in high-dose animals. Statistically significant changes in clinical chemistry were mostly observed in the high-dose groups. A number of statistically significant, but not dose-dependent, changes in terminal body weights and absolute and relative organ weights were reported (kidneys, liver, brain, spleen, thymus, lungs and cecum). The authors claim that the observed effects could be explained as physiological adaptive responses to excess carbohydrates, as reported by others, and that 20% of ribose induced few and minimal adverse effects of any kind. Analysis of microscopic histopathology revealed no evidence of treatment-related changes. A NOAEL of 3.6 and 4.4 g/kg bw per day was established in males and females, respectively.

Reference	Study	Dose and number in treatment group		Conclusion with regard to adverse effects
Griffiths et al. (2007a)	Oral embryotoxicity/teratogenicity study of D-ribose in female Wistar rats (OECD Test guideline 408, 1998) Study duration: 21 days	5%, 10% and 20% D-ribose in the diet. This represents a mean of 4.3, 7.9 and 9.9 g/kg bw per day in the low-dose, middle-dose and high-dose groups, respectively) (n = 20 M + 20 F)	Control diet with 0% supplemental D-ribose (n = 20 M + 20 F)	No statistically significant adverse effects were demonstrated on the developing embryo/fetus. The incidence of wavy ribs was statistically significantly higher in medium and high dose groups compared to control.

In a study of healthy thoroughbred racehorses, it was examined if dietary D-ribose supplementation would promote the formation of glycated proteins following exercise (Sinatra and Caiazzo, 2015). The background for the study was reference to the knowledge that non-enzymatic glycation of proteins by reducing sugars, such as glucose and D-ribose, may lead to formation of fructosamines and advanced glycation end products (AGEs), and that increased levels of AGEs following intraperitoneal injection of D-ribose in mice was associated with impaired cognitive function (Han et al., 2011). Two groups of 2-year-old horses were used, body weights (bw) ranged from 520 to 570 kg with a mean of 545 kg. One group (n = 5) received a daily dose of 30 g, the second group (n = 5) received a daily dose of 50 g (corresponding to a mean intake of 55 mg/kg body weight per day and 92 mg/kg bw per day, respectively). The ribose supplement was administered once daily, and doses were referred to be comparable to those used in humans (3-5 g per day used by exercising individuals). Study duration was 17 weeks, during which the horses were subjected to low-intensity exercises (8 weeks) followed by high-intensity exercises (9 weeks). Blood samples were collected and analysed for glycated plasma proteins at baseline and at the end of the two exercise regimens. The results showed that 17 weeks of ingestion of D-ribose at 30 or 50 g per day did not promote the formation of glycated plasma proteins in the racehorses after low-intensity or high-intensity exercise. No adverse effects of D-ribose supplementation were demonstrated. The authors concluded that supplementation with D-ribose was safe and did not cause glycation in this animal model.

In a subchronic study (13 weeks) by Griffiths et al. (2007b), groups of 5-week-old albino Wistar rats (20 males and 20 females per group) were administered diets 7 days/week in which 20% of the barley was replaced by the test article and/or pregelatinized potato starch containing 0, 5, 10 or 20% D-ribose (equivalent to mean daily intakes of 0.0, 3.6, 7.6 and 15.0 g/kg bw per day in males and 0.0, 4.4, 8.5 and 15.7 g/kg bw per day in females). Clinical observations were recorded once or twice a day. Body weights were recorded at day 0, weekly during the study and at sacrifice. Food consumption was measured daily. Water consumption was measured 4 or 5 consecutive days in week 1, 6 and 12. Ophthalmoscopic examinations were performed on all animals 5 days before the start of the study and on all control and high dose animals during the last week of the study. Haematological (haemoglobin, packed cell volume, prothrombine, neutrophils, lymphocytes), clinical chemistry (ALT, AST, total protein, albumin, albumin/globulin ratio, cholesterol, triglycerides, phopholipids) and urinalysis (water intake/24 h (fasted and non-fasted), urine volume/24 h (fasted and non-fasted) and urine density, urine glucose/24 h and urine glucose/creatinine ratio (all fasted)) measurements were taken at baseline and during the final week of the study on the same animals (10 per sex) from each dose group. Neurotoxicity screening was included and consisted of daily clinical observation, objective gross and histopathological analysis of the brain, spinal cord and peripheral nervous tissue and a functional observational battery (FOB) conducted during week 13 on the 10 males and 10 females not used for haematology, clinical chemistry and urinalysis measurements. At sacrifice, the organs of all animals were weighed and relative organ weights were calculated. Mean bw of all treated rats were decreased relative to those of controls. This effect was statistically significant in high-dose animals from day 7 onwards and in mid-dose females throughout most of the

study. In mid-dose males and low-dose males and females, significant decreases in weight were observed sporadically and less frequently. Mean bw of high-dose males at week 13 was 87% of controls, whereas mean bw of high-dose females was 91% of controls. Absolute and relative caecal weights were reported to be statistically significantly increased in a dose-dependent manner in both sexes (absolute weights at the two highest doses, relative weights at all doses). Absolute liver weight was reduced at the highest dose in males and increased at the low and mid-dose in females. Relative liver weight was increased in the mid-dose in males and at all doses in females, with the highest increase at the mid-dose. For kidneys, brain, spleen, thymus and lungs changes were not statistically significant and/or dose-dependent in either sexes. The authors concluded that most of these effects occurred as a physiological adaptive response to the high levels of carbohydrates in the diet, and that enlargement of the cecum and liver is commonly reported after high-level carbohydrate exposure. No significant differences were observed in the ophthalmoscopic examination. The haematological measurements showed sporadic, non-dose-responsive statistically significant effects and were considered unrelated to treatment. Cholesterol, triglyceride and phospholipid concentrations were significantly decreased in the high-dose males and non-significantly decreased in females. Most of the significant changes in clinical chemistry measurements and urinalysis were only observed in the high-dose treatment groups. At necropsy, no consistent macroscopic pathological findings were noted other than the yellow discoloration of abdominal fur (considered to be the result of excess urinary output). Microscopic evaluation of numerous tissues revealed no evidence of changes that could be attributed to treatment, as all findings were typical of this strain and age of rats and appeared to be randomly distributed among treated and control groups (data not shown). No treatment-related changes in hepatocellular morphology were observed upon staining of liver sections. Screening for neurotoxicity revealed no effects of treatment of D-ribose. In conclusion, the authors argued that it is scientifically reasonable to conclude that the present study supported a concentration of 5% D-ribose in the diet, corresponding to an average daily intake of 3.6 and 4.4 g/kg bw per day in male and female rats, respectively, as being the absolute no observed adverse effect level (NOAEL) for D-ribose.

In an oral embryotoxicity/teratogenicity study, female albino Wistar rats (28 rats/dose) were given 0, 5, 10 or 20% of D-ribose via the diet 7 days/week. This corresponds to a mean of 4.25 (individual range 3.6 – 4.6), 7.94 (7.0 – 8.7) and 9.91 (8.1 – 11.6) g/kg bw per day in the low-dose, middle-dose and high-dose groups, respectively (Griffiths et al., 2007a). The study was conducted concurrently with the study described above (Griffiths et al., 2007b). Exposure was from day 0 of gestation until maternal sacrifice on day 21. Body weights were recorded on day 0, 7, 14 and 21. Food consumption was assessed daily. After sacrifice on day 21, dams were examined for gross abnormalities and the organ weights were determined. Reproductive parameters were also assessed. Half of the foetuses in each litter were examined for soft tissue abnormalities and the remaining half were examined for skeletal abnormalities. In the control, 5, 10 and 20% groups 26, 26, 27 and 23 females, respectively, were confirmed pregnant. All animals survived to the Caesarean section on day 21 and clinical observations of the animals' appearance, general condition and behavior were reported to be unremarkable. Mean body weight gain and food consumption were

significantly reduced in the mid and high dose groups over the first two weeks of the study. However, during the last week of the study the mid and high dose groups had a significantly greater rate of weight gain compared to controls which resulted in no significant difference in body weight between the groups by the end of the study. Neither absolute nor relative liver weight was affected at any dose of D-ribose. Both absolute and relative caecal weights (both full and empty) were reported to be significantly increased at all dose levels when compared to control group. The enlargement of the cecum observed in the present study has been reported in laboratory rats after similar exposures. There was no difference in the gravid uterus weights or the carcass weights of the treated groups and the controls. Fecundity index, gestation index, pre-implantation loss, post-implantation loss and sex ratio of offspring were all unaffected by treatment with D-ribose. External observations of foetuses and placentas did not differ among study groups. No significant differences between treated and control groups in mean foetal and placental weights were seen. Observation of visceral abnormalities were unremarkable and did not differ between the control and treated groups. Statistically significant skeletal malformations were not observed in any of the study groups. There were no significant differences in the incidences of one or multiple wavy ribs in the control or test animals, but testing of the combined foetal and litter incidences revealed a statistically significantly higher incidence in the mid and high dose groups compared to control animals. A number of sporadic, non-dose-responsive variations in skeletal ossification were observed in all groups (data shown only for incomplete ossification of the frontal, parietal and interparietal bones). The authors refer to literature where historical control incidences for the noted skeletal anomalies (e.g. wavy ribs) and variations in ossification of the cranial bones have previously been reported and discussed for both the Wistar and other strains of rats. Both effects are generally described as reversible. Findings of delayed ossification have been reported to be very sensitive to gestational age, with skeletons at day 21 being very developed compared to skeletons from fetuses at day 19 and 20.

In conclusion, the result from the present study indicates that administration of D-ribose up to 20% of the diet during gestation days 0-21 induced few adverse effects on the dam or the developing embryo. The statistically significantly reduced, and then increased, weight gain in the mid and high-dose groups, and the statistically significant increase in caecal weight in the D-ribose groups compared to controls, may represent a metabolic challenge to maternal homeostatic mechanisms and thereby impact nutritional and endocrine communication between dam and subsequent offspring. Thus, a clear NOAEL derived for developmental toxicity is 5% D-ribose in the diet, corresponding to an average intake of D-ribose between 3.6 and 4.6 g/kg bw per day.

2.4.2.1 Interactions

There was no information concerning interactions in the literature reviewed in the present risk assessment. The absence of information in the selected literature does not document an absence of interactions.

2.4.2.2 Allergic sensitisation (including adjuvant effects)

There was no information concerning allergic sensitisation or allergy adjuvant effects in the literature reviewed in the present risk assessment. The absence of information in the selected literature does not document an absence of allergic sensitisation or allergy adjuvant effects.

2.4.3 Mutation and genotoxicity studies

Four unpublished and two published mutagenicity and genotoxicity studies on D-ribose are referred to in the "Application for the approval of D-ribose for use as an ingredient in foods, food supplements and food for particular uses" (Bioenergy Life Science Inc., 2008) and summarised in the "Draft initial opinion on an application under the novel foods regulation for D-ribose"(Advisory committee on novel foods and processes, 2016). Three bacterial reverse mutation tests were referred to. One study (unpublished) used the *S. typhimurium* strains TA 1535, TA 1538, TA 98 and TA 100 as well as the *E. coli* strain WP2 *uvr*A, a second study (Wilmer et al., 1981) used *S. typhimurium* strains TA 100 and TA 98, and a third study (Aiyar and Subba, 1977) used *S. typhimurium* 434. Other tests referred are one *in vitro* chromosomal aberration assay in Chinese hamster ovary cells, one gene mutation assay at the thymidine kinase locus of mouse lymphoma L5178Y cells and one *in vivo* rat bone marrow micronucleus assay. All results from the mutagenicity and genotoxicity tests were negative, indicating that D-ribose is not genotoxic in bacterial or mammalian cells.

2.4.4 Vulnerable groups

No specific vulnerable groups were identified with respect to adverse health effects due to intake of D-ribose from food supplements. However, oral intake of D-ribose can be associated with a decrease in blood glucose levels. For example, asymptotic and transient hypoglycemic effects were observed after bolus doses of 10 g D-ribose or greater. In these studies, subjects were given D-ribose formulations containing no other energy source following an overnight fast (Bioenergy Life Science Inc., 2008). There was no further information available regarding potential effects of D-ribose on diabetic persons.

No information was found regarding effects of daily intake of D-ribose in pregnant and lactating women.

2.5 Summary of hazard identification and characterisation

D-ribose can be synthesized by almost every tissue in the body from carbohydrates such as glucose. D-ribose is a component of the genetic material RNA. It is also a structural component of ATP, the primary source of cellular energy. D-ribose is available in small amounts in the diet via ripe fruits and vegetables (Dhanoa and Housner, 2007). Orally administered D-ribose is generally thought to be absorbed in the small intestine by passive diffusion.

No serious adverse health effects of D-ribose were identified at the doses (10-20 g/day) reported in the human studies included in this opinion. However, a mild and not statistically significant state of hypoglycemia and hyperuricemia could be observed after oral consumption of 20 g/day (Seifert et al., 2008).

Three animal studies are included in the hazard characterisation. In one study performed in race horses, no adverse effects were observed after exposures to 30 and 50 g/day (corresponding to 55 mg/kg body weight per day and 92 mg/kg bw per day, respectively). In one subchronic oral toxicity study, rats were given 5%, 10% and 20% of D-ribose in the diet (3.6, 7.6 and 15.0 g/kg bw per day in males and 4.4, 8.5 and 15.7 g/kg bw per day in females) (Griffiths et al., 2007a; Griffiths et al., 2007b). NOAELs of 3.6 and 4.4 g/kg bw per day (lowest dose) in male and female rats were found. This was based on a statistically significant decreases in body weight in the two higher doses and changes in clinical chemistry (mostly at the highest dose). In another study in rats given 5%, 10% and 20% of D-ribose in the diet, the NOAEL for embryo toxicity/teratogenicity of D-ribose was the 5% concentration, corresponding to an average intake of 3.6 and 4.6 g/kg bw per day in males and females, respectively. This NOAEL was primarily based on a statistically significant higher incidence of one or multiple wavy ribs in the mid- and high-dose groups compared to control animals.

For risk assessments of macronutrients (i.e. fat, carbohydrate, protein or their substitutes) nutritional as well as toxicological aspects may be considered (Borzelleca, 1996; Dybing et al., 2002; Munro et al., 1996). In the present risk assessment, potential nutritional effects of D-ribose were not specifically evaluated.

For the risk characterization, the intake of 20 g/day of D-ribose (corresponding to 286 mg/kg bw per day in a 70 kg adult) considered to be without appreciable health risk in human adults, and a NOAEL of 3.6 g/kg bw per day (from a subchronic toxicity and embryotoxicity/teratogenicity study in rats) were used for comparison with the estimated exposure from food supplements.

3 Exposure / Intake

3.1 Food supplements

Exposure of D-ribose was estimated from the intake of food supplements. The intake of D-ribose was estimated for the age groups children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years).

NFSA requested VKM to perform a risk assessment of 3100 and 6200 mg/day of D-ribose in food supplements for children (10 to <14 years), adolescents (14 to <18 years) and adults. The default body weights (bw) for these groups as determined by EFSA were used: 10 to <14 years; 43.4 kg, 14 to <18 years; 61.3 kg and adults (≥18 years); 70.0 kg (EFSA, 2012). From the daily doses of 3100 mg and 6200 mg, the estimated exposures per kg body weight to D- ribose from food supplements for the various age groups is presented in Table 3.1-1.

Table 3.1-1   Estimated exposure to D-ribose in children, adolescents and adults from food supplements.

Age groups	Daily doses (mg)	Body weight (kg)	Exposures (mg/kg bw per day)
Children (10 to <14 years)	3100	43.4	71.4
	6200	43.4	142.9
Adolescents (14 to <18 years)	3100	61.3	50.6
	6200	61.3	101.1
Adults (≥18 years)	3100	70.0	44.3
	6200	70.0	88.6

3.2 Other sources

D-ribose is available in small amounts in the diet via ripe fruits and vegetables (Dhanoa and Housner, 2007). D-ribose is used as an ingredient in some so-called energy drinks. In the EU, D-ribose can be used in cosmetics as skin conditioner and humectant (CosIng, 2015).

4 Risk characterisation

NFSA requested VKM to perform a risk assessment of doses of 3100 mg/day and 6200 mg/day of D-ribose in food supplements for the general population, ages 10 years and above. In this opinion, VKM has used the intake of 20 g/day of D-ribose (corresponding to 286 mg/kg bw per day) from the human studies and a NOAEL of 3.6 g/kg bw per day from a study in rats for comparison with the estimated exposure.

Human data

No serious adverse health effects were identified at the doses (10 - 20 g per day) reported in the human studies included in this opinion. The doses of 3100 mg/day or 6200 mg/day of D-ribose from food supplements is below the level of 20 g/day in all age groups. The doses are also within the range of endogenous synthesis of D-ribose which has been estimated to be from 2.7 g per day (women) to 16.5 g per day (men).

No studies on children (10 to <14 years) and adolescents (14 to <18 years) were identified. Based on the included literature there was no evidence indicating that age affects tolerance for D-ribose. Therefore, in this risk characterisation a tolerance as for adults, based on body weight, were assumed for these age groups.

Animal data

The NOAEL used for the risk characterisation of D-ribose is 3.6 g/kg bw per day from the 13 weeks subchronic toxicity and embryotoxicity/teratogenicity studies in rats. This value is used to calculate the Margin of Exposure (MOE), the ratio of the NOAEL to the exposure.

An acceptable MOE value for a NOAEL-based assessment of chemicals based on an animal study is ≥100, which includes a factor 10 for extrapolation from animals to humans and a factor 10 for interindividual human variation (EPA, 2012). A MOE below 100 may also be acceptable; however, such assessments must be based on supporting scientific literature and expert judgement.

The calculated margins between the NOAEL of 3.6 g/kg bw per day from the rat study and the exposure of D-ribose from food supplements (MOE values) are presented in Table 4.1-1. The estimated exposures are presented in Table 3.1-1.

Table 4.1-1 The calculated margins between the NOAEL from a rat study and the exposure to D-ribose from food supplements (MOE values) for the various age groups.

Age groups	3100 mg/day	6200 mg/day

Age groups	3100 mg/day	6200 mg/day
Children (10 to <14 years)	50.4	25.2
Adolescents (14 to <18 years)	71.1	35.6
Adults (≥18 years)	81.3	40.6

The calculated MOE values ranged from 25.2 to 81.3 for a daily intake of 3100 mg/day or 6200 mg/day of D-ribose. In this case, MOE values below 100 are regarded as acceptable since D-ribose is present in all cells in the body and the daily doses from food supplements are in the same order as the endogenous production, which ranges from 2.7 g/day (women) to 16.5 g/day (men) (Bioenergy Life Science Inc., 2008). Based on human data and the calculated MOE values from the animal study, VKM concludes that it is unlikely that daily doses of 3100 mg or 6200 mg D-ribose in food supplements causes adverse effects in children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years).

5 Uncertainties

5.1 Uncertainty in hazard identification and characterisation

Several of the studies referred to are randomized control trials (RCTs), specifically designed to investigate the positive effects of D-ribose, and not negative effects. Some mild negative effects are reported based on self-reporting questionnaires, and also some biomarkers for physiological changes that may not necessarily represent negative effects, e.g. blood glucose levels, were reported.

No studies on children and adolescents were identified. A tolerance as for adults, based on body weight, was assumed for these groups.

In the present risk assessment, potential negative nutritional effects of D-ribose were not specifically evaluated.

5.2 Uncertainty in exposure

With use of the default (mean) body weight of an age (population) group, the variance in all individuals in the group will not be covered.

5.3 Uncertainty in risk characterisation

For risk assessments of macronutrients (i.e. fat, carbohydrate, protein or their substitutes) nutritional as well as toxicological aspects may be considered. When macronutrients are fed to rodents at high dietary levels, nutritional imbalance and physiological effects may lead to secondary adverse effects.

6 Conclusions with answers to the terms of reference

The Norwegian Scientific Committee for Food Safety (VKM) has, at the request of the Norwegian Food Safety Authority, assessed the risk of D-ribose (3100 mg/day and 6200 mg/day) in food supplements. The present risk assessment is based on previous risk assessments and a literature search.

No serious adverse health effects were identified at the dose range of 10 - 20 g/day reported in the human studies included in this opinion.

No human studies on children (10 to <14 years) and adolescents (14 to <18 years) were identified. Based on the included literature there was no evidence indicating that age affects tolerance for D-ribose. Therefore, in this risk characterisation a tolerance as for adults, based on body weight, were assumed for these age groups.

The NOAEL of 3.6 g/kg bw per day from a 13 week subchronic toxicity and embryotoxicity/teratogenicity studies in rats was used to calculate the MOE. The calculated MOE values from the rat study for a daily intake of 3100 mg/day were 50.4, 71.1 and 81.3 for children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years), respectively. The calculated MOE values for a daily intake of 6200 mg/day were 25.2, 35.6 and 40.6 for children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years), respectively. In this case, MOE values below 100 are regarded as acceptable since D-ribose is present in all cells in the body and the daily doses from food supplements are in the same order as the endogenous production, which ranges from 2.7 g per day (women) to 16.5 g per day (men) (Bioenergy Life Science Inc., 2008).

VKM concludes that it is unlikely that daily doses of 3100 mg or 6200 mg D-ribose in food supplements causes adverse effects in children (10 to <14 years), adolescents (14 to <18 years) and adults (≥18 years).

An overview of the conclusions is presented in Table 6.1. Estimated exposures unlikely to cause adverse health effects (below the values for comparison) are shown in green.

Table 6.1   An overview of the conclusions on daily intake of D-ribose from food supplements. Green: estimated exposure to D-ribose is unlikely to cause adverse health effects.

Food supplement	3100 mg/day	6200 mg/day
Children **(10 to <14 years)**		
Adolescents **(14 to <18 years)**		
Adults **(≥18 years)**		

7 Data gaps

There is a lack of human studies that have investigated the effect of high doses for longer periods than 8 weeks .

No studies on adverse health effects of D-ribose in children, adolescents, pregnant women or lactating women were identified.

There is a lack of chronic toxicity studies in animals.

8 References

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9 Appendix

Database: Embase <1974 to 2016 January 07>, Ovid MEDLINE(R) In-Process & Other Non-Indexed Citations, Ovid MEDLINE(R) Daily, Ovid MEDLINE(R) and Ovid OLDMEDLINE(R) <1946 to Present>

Search Strategy:

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1. ribose*.ti. (10954)
2. (risk* or safety or adverse or side-effect*1 or hazard* or harm* or eosinophil* or negative or contraindicat* or contra-indicat* or interact* or toxicity or toxic).tw. (9804998)
3. 1 and 2 (2072)
4. (conference abstract* or letter* or editorial*).pt. (4883060)
5. 3 not 4 (1941)
6. limit 5 to (danish or english or norwegian or swedish) (1907)
7. remove duplicates from 6 (1016)
8. d-ribose*.ti. (48)