



**Vitenskapskomiteen for mattrygghet**  
Norwegian Scientific Committee for Food Safety

# Comparison of organic and conventional food and food production

## Part III: Human health – an evaluation of human studies, animal models studies and biomarker studies

### Opinion of the Panel on Nutrition, Dietetic Products, Novel Food and Allergy of the Norwegian Scientific Committee for Food Safety

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# **Organic food and human health – an evaluation of human studies, animal model studies and biomarker studies**

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## Acknowledgements

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## Summary

The Panel on Nutrition, Dietetic Products, Novel Food and Allergy has evaluated and summarised current scientific knowledge about organic food consumption and impact on human health. The aim was to investigate whether consumption of organically produced versus conventionally produced foods has a positive and/or negative influence on human health. A specification of the substances which may be connected to the reported health effects are outlined where possible. Comments have been included for differences in levels (compared to conventional food) of nutrients and other bioactive compounds.

The evaluation is based on comprehensive literature searches. However, few relevant studies were found. Studies with humans (9 papers), animal model studies (9 papers) and biomarker studies (13 papers) including one or more health outcomes were included. Studies only describing levels of various nutrients or other bioactive compounds in conventional foods versus organic foods were not considered. Only studies where organic food production was similar to the practises embedded in the European member state regulation were included.

The following clinical health outcomes were investigated in the included human studies: Atopy, eczema, respiratory disease, semen quality, hypospadias and risk factors for cardiovascular disease. The studies are so far scarce and have several limitations, and the clinical health outcomes studied have been investigated in too few studies to allow for conclusions. No conclusion could therefore be drawn regarding semen quality, hypospadias or risk factors for cardiovascular diseases. For immune-associated outcomes, there are some indications that organic food consumption may have a more beneficial effect than conventionally produced food.

None of the included studies on human health reported negative health effects from organic food consumption compared with conventional food. However, the studies were not specifically designed to investigate safety.

In the nine included animal studies, various general health parameters were tested, but all studies included some immune parameters. Animals studied were rats (six studies), mice, chicken and *Drosophila Melanogaster* (fruit flies) (one study each). Results from these studies might indicate a positive effect on some outcomes; however, the relevance of extrapolating from animal studies to humans remains a challenge. Organically produced plant foods in some well performed animal studies showed a positive effect on animal physiology which may have an impact on animal health, i.e. immune parameters, hormonal balance and activity level. Not all studies substantiated these findings. Possible explanations for the discrepancies might be found in the differences in study design, including animal breed, plant food used, growing seasons and geographical location.

Thirteen included papers investigated changes in biomarkers of antioxidant capacity, specific fatty acids, copper and zinc bioavailability and pesticides from consumption of organic foods compared with conventional foods. The study designs and biomarkers measured in these investigations varied.

The overall impression of the biomarker studies in humans is that there is probably no difference in antioxidant capacity, after absorption, between organic and conventional foods, but better designed studies are needed. There are some indications of increased concentrations of anti-inflammatory and growth stimulating trans fatty acids in human milk from mothers using predominantly organically produced dairy and meat products. In the USA, a lower urinary concentration of pesticides was found in children consuming an organic diet compared to a conventional diet. It was, however, difficult to relate any biomarkers to better health or protection against disease.

Our conclusions are in line with several previous reviews.

There were several methodological challenges in studies investigating organic food and the effect on human health. A frequent problem was inadequate descriptions of the foods which were tested in the various studies, also in terms of production conditions. Another major problem was the lack of a suitable design, both for experimental studies, but perhaps even more so for clinical trials. The included animal studies were well-designed and investigated relevant endpoints and selection of biomarkers.

The present report has identified a marked need for more and improved studies. Research within the field shows lack of consistent findings and inconclusive results. It is of importance to gain more knowledge both on eventual positive and negative health effects.

## Norsk sammendrag

Faggruppen for ernæring, dietetiske produkter, ny mat og allergi har vurdert og sammenfattet vitenskapelige kunnskap om sammenhenger mellom inntak av økologisk mat og human helse. Målet har vært å undersøke om inntak av økologisk produsert versus konvensjonelt produsert mat har en positiv og / eller negativ innvirkning på menneskers helse. En spesifisering av eventuelle stoffer i maten som kan knyttes til de rapporterte helseeffekter er angitt der det er mulig. Det er også gitt kommentarer til enkelte forskjeller i nivåer (sammenliknet med konvensjonell mat) av næringsstoffer og andre bioaktive komponenter.

Evalueringen er basert på omfattende litteratursøk. Imidlertid ble det funnet få relevante studier. Studier som hadde undersøkt helseutfall og sammenliknet økologisk mat med konvensjonell mat er inkludert; humanstudier (9 artikler), dyremodellstudier (9 artikler) og biomarkørstudier (13 artikler). Studier som bare beskrev nivåer av ulike næringsstoffer eller andre bioaktive komponenter i konvensjonelle matvarer versus økologiske matvarer ble ikke inkludert. Kun studier av økologisk matproduksjon som kan sammenliknes med økologisk produksjon i henhold til EUs reguleringer ble inkludert.

Følgende kliniske helseutfall er undersøkt i de inkluderte humanstudiene: Atopi, eksem, luftveissykdom, sæd kvalitet, hypospadi og risikofaktorer for hjerte- og karsykdommer. Til nå er det få studier som har sammenliknet helseutfall fra økologisk i forhold til konvensjonell mat, og de inkluderte studiene har flere begrensninger. Det kan derfor ikke gis noen konklusjon for helseutfall som sæd kvalitet, hypospadi eller risikofaktor for hjerte- og karsykdommer. For immunassosierte helseutfall, er det noen indikasjoner på at inntak av økologiske mat kan ha en mer gunstig effekt enn konvensjonelt produsert mat.

Ingen av de inkluderte humanstudiene rapporterte negative helseeffekter fra økologisk mat sammenliknet med konvensjonell mat. Imidlertid var ikke studiene særskilt designet for å undersøke risiko og mattrygghet.

I de ni inkluderte dyrestudiene er ulike generelle helseparametere undersøkt, og alle studiene inkluderte immunparametere. Dyreartene som ble undersøkt inkluderte rotter (seks studier), mus, kylling og *Drosophila Melanogaster* (bananflue) (én studie for hver art). Resultatene fra disse studiene kan indikere en positiv effekt på enkelte utfall hos dyrene. Økologisk produsert planteføde i noen godt utførte dyrestudier viste en positiv effekt på immunparametere, hormonbalansen og aktivitetsnivå hos dyrene. Imidlertid bekreftet ikke alle studiene disse funnene. Mulige forklaringer på divergerende resultater kan skyldes forskjeller i studiedesign, dyreart, planteføde som ble gitt, vekstsesong og geografisk beliggenhet. Relevansen av å ekstrapolere fra dyreforsøk til mennesker er imidlertid stadig en utfordring.

I de 13 inkluderte biomarkørstudiene ble det undersøkt endringer i antioksidant kapasitet, særskilte fettsyrer, biotilgjengelighet av kobber og sink og plantevernmidler i forbindelse med inntak av økologisk, sammenliknet med konvensjonelle mat. Studiedesignet i de ulike biomarkørstudiene varierte.

Det overordnede inntrykket fra biomarkørstudiene er at det trolig ikke er noen forskjell i antioksidantkapasitet etter inntak og absorpsjon av økologisk versus konvensjonelt dyrket mat, og det er behov for flere og bedre studier. Det er noen indikasjoner på økte konsentrasjoner av antiinflammatoriske og vekststimulerende transfettsyrer i morsmelk fra mødre som bruker hovedsakelig økologisk produserte meieri og kjøttprodukter. Det ble funnet lavere konsentrasjoner av pesticider i urin hos amerikanske barn som spiste økologisk mat når man sammenliknet med barn som spiste konvensjonelt produsert mat. Det er imidlertid vanskelig å si noe om hvorvidt disse endringene i biomarkører vil ha positiv effekt på helsen eller beskytte mot sykdom.

Det er rapportert om enkelte forskjeller i konsentrasjoner av næringsstoffer og andre bioaktive komponenter i økologisk produsert mat sammenliknet med konvensjonelt produsert mat. Om disse forskjellene vil ha noen betydning for helsen er imidlertid ikke vist.

VKMs konklusjoner er i tråd med flere tidligere publiserte vitenskapelige oversiktsartikler.

Det er flere metodiske utfordringer i de inkluderte humanstudiene. Eksempelvis var det mangelfulle beskrivelser av de matvarene som ble spist i de ulike studiene, og forholdene rundt produksjon var ikke alltid tilstrekkelig dokumentert. En annen svakhet var kvaliteten på selve studiedesignet for de eksperimentelle studiene, men kanskje særlig for de kliniske studiene. De inkluderte dyrestudier hadde god kvalitet og undersøkte relevante biomarkører og helseutfall.

Denne VKM-vurderingen har avdekket et stort behov for flere og bedre studier. Forskning innen feltet viser mangel på konsistente funn og sprikende resultater. Det er viktig å få mer kunnskap både om eventuelle positive og negative helseeffekter fra økologisk mat.

## Abbreviations

ADI; Acceptable daily intake  
BMI; Body mass index  
CLA; Conjugated linoleic acid isomers  
CVD; Cardiovascular disease  
eTBARS; Erythrocyte thiobarbituric acid reactive substance  
FFQ; Food frequency questionnaire  
GMO; Genetically modified organism  
KLH; Keyhole limpet haemocyanine  
OFA; Danish Organic Farmers' Association  
ORAC; The oxygen radical absorbing capacity  
PEM; Protein-energy-malnourished  
RCT; Randomised controlled trials  
SD; Standard deviation  
p-tHcy; plasma total homocysteine  
TEAC; trolox equivalent antioxidant capacity  
Ø-label; Debio's certification label

## Definitions

### **Biodynamic agriculture**

Biodynamic agriculture is a method of organic farming originally developed by Rudolf Steiner (Austrian philosopher 1861 – 1925) that employs what proponents describe as "a holistic understanding of agricultural processes". Being one of the first sustainable agriculture movements, it treats soil fertility, plant growth, and livestock care as ecologically interrelated tasks, emphasising planetary impact on plants and animals. All biodynamic farms in Norway comply with the regulations for organic farming and are certified and inspected by Debio. In addition to the Ø-label, biodynamic products are marked with the international Demeter-sign.

### **Conventional farming**

In the present report conventional farming and conventional products include all which are not defined as organic agriculture.

### **Debio**

The Norwegian certification organisation for organic products. Most of Debio's services deal with the inspection of organic production in accordance with the Norwegian "Regulations on the Production and Labelling of Organic Agricultural Products". The inspection services are based on an agreement with the Norwegian Food Safety Authority, and the regulation is based on the EU Council Regulation 2092/91. It covers farming, processing, import and marketing of organic agricultural products in Norway.

### **Human health**

In the present report we have investigated organic foods in relation to various health outcomes, be it altered incidence of some health condition or effects on biomarkers of health and disease. For the purpose of this report "human health" in the context of organic food is therefore defined as "The organic diet's or diet constituent's ability to influence indicators of health or disease".

**Organic farming**

Modified from Debio and the Norwegian Food Safety Authority (Mattilsynet, NFSA): The term “organic” is protected by law for food and feed products and can only be used when certified by Debio or a similar foreign control body. The Ø-label is therefore a guarantee for the product being organically produced. Debio inspects the farms and operators engaged with processing, packaging, import and trade of organic foods. Norwegian regulations for organic food and feed production are almost identical to the parallel European regulations. Foods from organic farms are produced with use of animal manure and other organic fertilisers; the use of chemical-synthetic pesticides is forbidden. There are restrictions on the use of preservatives and the use of artificial aroma- and colours are not allowed. All use of genetically modified organisms (GMO) is also prohibited. (

From The International Federation of Organic Agricultural Movements: Organic agriculture is a production system that sustains the health of soils, ecosystems and people. It relies on ecological processes, biodiversity and cycles adapted to local conditions, rather than the use of inputs with adverse effects. Organic agriculture combines tradition, innovation and science to benefit the shared environment and promote fair relationships and a good quality of life for all involved.

**EU regulation of organic production:** Council Regulation (EC) No 834/2007 of 28 June 2007 on organic production and labelling of organic products and repealing Regulation (EEC) No 2092/91. The EEA countries (Norway, Iceland and Lichtenstein) are in the process to implement Regulation (EC) No 834/2007 and its appurtenant regulations.

**Norwegian regulations:** The Norwegian Food Safety Authority has implemented the Norwegian regulations in «Forskrift om økologisk produksjon og merking av økologiske landbruksprodukter og næringsmidler» (<http://lovdata.no/dokument/SF/forskrift/2005-10-04-1103>).

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## Background

The goal of the Norwegian government is that 15% of the agricultural production is organic in 2020 (St. Meld. 9, 2011-2012). However, knowledge on the impact of an increase in organic production in Norway is limited. If and how organic production practices may affect human health, animal health and welfare, plant health, the environment and sustainability is not clear.

In order to be able to give scientifically based information and advice on this issue to consumers and other target groups, the Norwegian Food Safety Authority (NFSA) requested a scientific evaluation of current research and other data on organic food and food production from The Norwegian Scientific Committee for Food (VKM). The scientific evaluation and the knowledge will also be used in connection with the NFSA's regulatory and international work on organic food production. The NFSA first prepared a draft request that was put out for public consultation. Remarks from the bodies that commented on the proposal clearly stated that there are limitations in the basic data for such an evaluation. NFSA therefore limited the scope and focus of the request somewhat. Sustainability aspects and environmental impact of organic and conventional agricultural practices are not addressed. In addition, organic aquaculture, which has only been practiced for a few years, is excluded from the request.

All foodstuffs on the market shall be safe and wholesome. Whereas all food produced and marketed shall comply with relevant legislation, food marketed as organic must in addition comply with regulations specific for organic production.

**Organic food production** is defined in Council Regulation (EC) No 834/2007 of 28 June 2007 on organic production and labelling of organic products as "The use of the production method compliant with the rules established in this Regulation, at all stages of production, preparation and distribution". The regulation on organic food production is part of the EEA Agreement and covers inputs, crop production, livestock production, rules for processing, labeling, and inspection, and provides provisions for imports from third countries.

According to Council Regulation (EC) No 834/2007, organic production shall be based on the following principles (article 4):

- (a) the appropriate design and management of biological processes based on ecological systems using natural resources which are internal to the system by methods that:
  - i) use living organisms and mechanical production methods;
  - ii) practice land-related crop cultivation and livestock production or practice aquaculture which complies with the principle of sustainable exploitation of fisheries;
  - iii) exclude the use of GMOs and products produced from or by GMOs with the exception of veterinary medicinal products;
  - iv) are based on risk assessment, and the use of precautionary and preventive measures, when appropriate;
- (b) the restriction of the use of external inputs. Where external inputs are required or the appropriate management practices and methods referred to in paragraph (a) do not exist, these shall be limited to:
  - i) inputs from organic production;
  - ii) natural or naturally-derived substances;
  - iii) low solubility mineral fertilisers;
- (c) the strict limitation of the use of chemically synthesised inputs to exceptional cases these being:

- i) where the appropriate management practices do not exist; and
- ii) the external inputs referred to in paragraph (b) are not available on the market; or
- iii) where the use of external inputs referred to in paragraph (b) contributes to unacceptable environmental impacts;

(d) the adaptation, where necessary, and within the framework of this Regulation, of the rules of organic production taking account of sanitary status, regional differences in climate and local conditions, stages of development and specific husbandry practices.

## Terms of reference

The Norwegian Food Safety Authority (NFSA) requests the Norwegian Scientific Committee for Food Safety (VKM) to evaluate current scientific knowledge of organic production and organically produced food based on existing national and international research results and other documentation. The NFSA wants the evaluation to focus primarily on Norwegian production.

NFSA has found it appropriate to divide this comprehensive evaluation of organic production and organic food into five parts:

1. Plant health – plant production
2. Animal health – animal welfare and feed
3. Human health – nutrition and contaminants
4. Human health – hygiene and pathogens
5. Human health – pesticide residues

NFSA would like VKM to compare the effects of organic versus conventional production based on the evaluations that are done in the five areas above. If lack of data prevents such a comparison, this should also be reported.

### **Part III, IV and V. Human health**

NFSA requests VKM to evaluate the impact on human health in Norway of eating organic versus conventionally produced food. The assessment is for practical reasons divided into three parts.

#### **Part III. Human health - nutrition and contaminants**

NFSA requests VKM to identify and/or assess:

- if consumption of organic food versus conventional food has a positive and/or negative influence on human health? Specification of substances that may be connected to reported health effects is wanted and also possible differences in levels of these substances (between conventional and organic products).

# 1 Introduction

The Norwegian Scientific Committee for Food Safety (Vitenskapskomiteen for mattrygghet, VKM) has at the request of the Norwegian Food Safety Authority (Mattilsynet, NFSA) compared organic and conventional food and food production in relation to possible impact on plant health, animal health and welfare and human health. The assessment is based on published peer reviewed scientific literature and assessment reports from international and national scientific bodies.

The following aspects of organic food production were not addressed in the assessment as they were not part of the request; sustainability aspects and environmental impacts of organic and conventional agricultural practices, and furthermore: aquaculture, because organic aquaculture has only been practiced for a few years.

At the request of the Norwegian Food Safety Authority the assessment was divided into five parts addressing:

- I) Plant health and plant production (assessed by Panel on Plant Health)
- II) Animal health and animal welfare (assesses by Panel on Animal Health and Welfare)
- III) Humane health - nutrition and contaminants (Panel on Nutrition, Dietetic Products, Novel Food and Allergy)
- IV) Human health – hygiene and pathogens (assessed by Panel on Biological Hazards)
- V) Pesticide residues (assessed by Panel on Plant Protection Products)

The present report focuses solely on current scientific knowledge about organic food consumption and impact on human health. VKM appointed a working group consisting of VKM members and one external expert to prepare a draft opinion. The opinion was approved by VKMs Panel on Nutrition, Dietetic Products, Novel Food and Allergy. The Scientific Steering Committee of VKM approved the final opinion, i.e. this document.

## 2 Literature search

### 2.1 SEARCH STRATEGY

Two literature searches were performed to retrieve publications addressing putative health effects of organically compared with conventionally produced foodstuffs, and only outcomes of relevance to human health were included. The strategy for the literature searches was discussed and accepted in the project group and set up together with a librarian. Test searches were conducted to find relevant terms, search words and controlled vocabulary (MeSH and EMTREE). Literature search including different terms for organic and organic foods in the titles was conducted in Medline, Embase and ISI Web of Science. A second search was conducted to capture studies in subjects with an anthroposophic lifestyle. The latter search included several specific terms for health outcomes and was also conducted in Medline, Embase and ISI Web of Science. The databases were used to ensure comprehensive study retrieval. The searches were conducted using a combination of both controlled vocabulary and text word searching as shown in Appendix 1.

The searches were limited by omitting abstracts and by limiting the languages to Danish, English, French, German, Norwegian and Swedish. Since a common regulatory framework for organic production was introduced in all member states of the European Economic Area in 1991, the search period was limited to the years 1991 until present. In addition, in the first

search the term organic\* or biodynamic\* was required in the title. The second search was conducted to capture studies of populations with a lifestyle including an organic diet. The first search was performed on the 17<sup>th</sup> of October 2013; the second search was performed on the 29<sup>th</sup> of October 2013.

## 2.2 PUBLICATION SELECTION

We considered four main study types for inclusion in this report: human studies, biomarker studies, animal studies, in addition to other relevant studies. The criteria for inclusion of a study were:

- It must be a full-text article
- It must be published and peer-reviewed
- It must contain a health outcome<sup>1</sup> (endpoint) that is related to organic food
- Organic production from countries outside the European Economic Area must be certified by an approved body
- Must include a comparison of organic and conventional foods

Studies only describing levels of various nutrients or other bioactive compounds in conventional foods versus organic foods were therefore not included. The first literature search identified 816 articles (Figure 2.1), of which 65 articles were included as potentially relevant (including reviews). The examination of the full text versions resulted in the exclusion of 23 studies which did not meet the inclusion criteria. Only one new paper was retrieved in the second literature search. The first assessment was performed by two reviewers for all articles and the second assessment was performed by the reviewers separately. A manual search of reference lists identified an additional one relevant review that was included.

A final total of 44 publications; 9 epidemiological/clinical studies, 9 animal studies, 13 biomarker studies, 1 other relevant study and 12 reviews were identified and included in this report (see Figure 2.1). For all included single studies, relevant data were extracted and entered into summary tables (Appendix 2). The data extraction was performed separately by the reviewers.

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<sup>1</sup>A definition of *human health* for the purpose of this assessment is given on page 8.

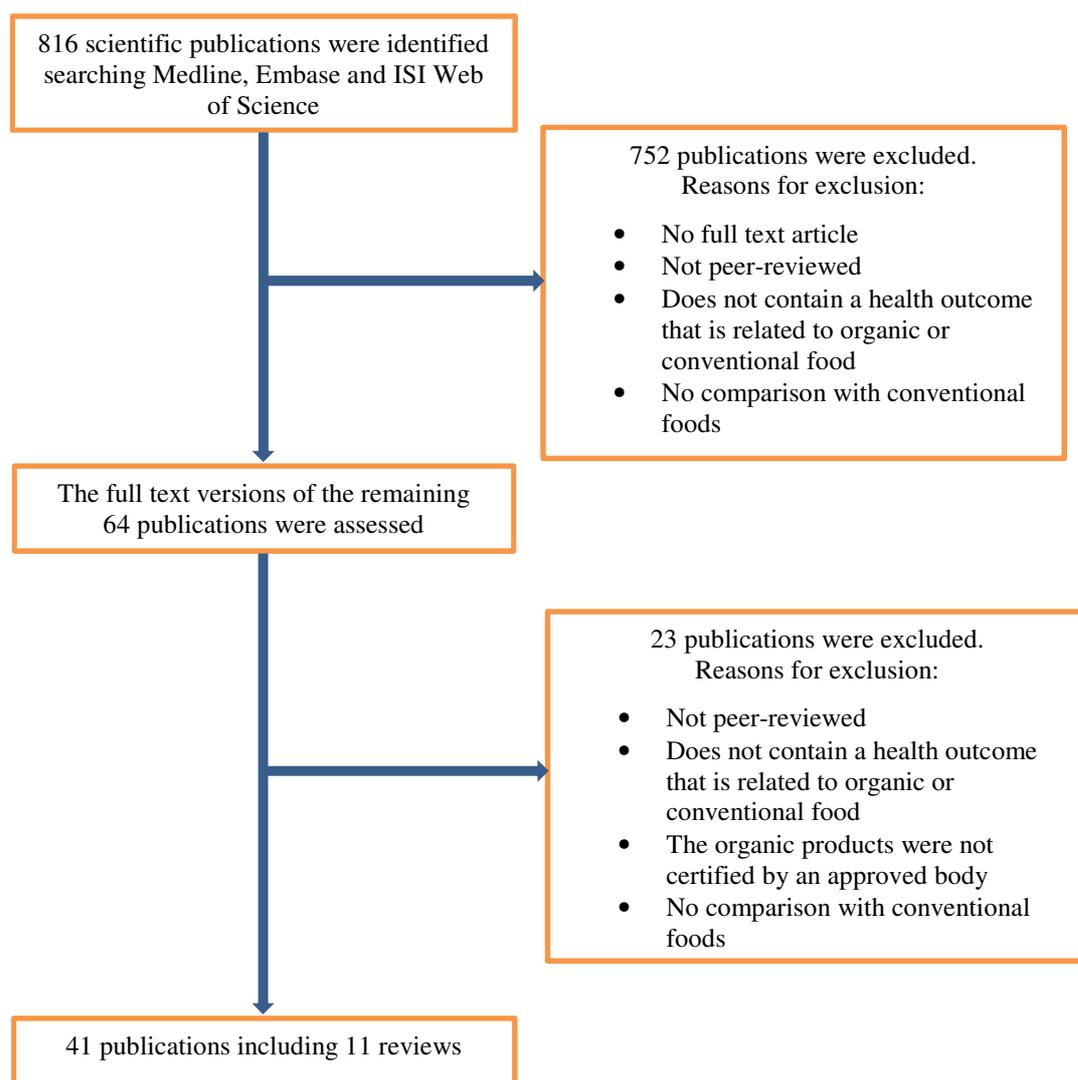


Figure 2.1: Flowchart; the first literature search and the selection of publications

### 3 Results and discussion

In the present assessment, the following categories of studies related to human health were included:

- Human studies including randomised and non-randomised controlled trials, or observational studies (prospective, case-control or cross-sectional) which included health endpoints of various kinds.
- Animal studies where animal models were explicitly used as a proxy for human physiological, biochemical or other hypothesised mechanisms in humans.
- Human studies using biomarkers of health as a proxy for health effects.
- *In vitro* and *ex vivo* studies in human or animal cell lines and serum used to investigate human-related cell mechanisms.

### 3.1 EPIDEMIOLOGICAL/CLINICAL STUDIES IN HUMANS

The literature search resulted in articles with the following clinical health outcomes in relation to organic food use in humans: Atopy, eczema, respiratory disease, sperm quality, hypospadias and risk factors for cardiovascular disease. One study compared children with an anthroposophic lifestyle including organic food with children from homes using conventional produced food. Hay fever and asthma symptoms were studied among organic and conventional farms. One study of atopic manifestation among infants brought up with respectively organic and conventional food was included. Four papers were identified investigating organic farmers and/or use of organic food in relation to semen quality. Organic food intake and exposure to pesticides were evaluated in two papers. A case control study was included where the frequency use of organic food by the mothers during pregnancy was analysed in relation to hypospadias in the children. One study investigated nutritional status and risk factors for cardiovascular disease with use of Italian Mediterranean organic diet. Table 3.1 gives an overview of all the studies in humans included in this opinion.

**Table 3.1: Overview of all the studies in humans included in this opinion.**

Authors	Country	Study design	Number of participants	Study characteristic	Main endpoint	Main findings
Alm <i>et al.</i> (1999)	Sweden	Cross-sectional	295 Steiner school children, 380 controls	Anthroposophic lifestyle	Atopy	OR (95% C.I.) for atopy was 0.63 (0.43,0.91) for the Steiner school children.
Kummeling <i>et al.</i> (2008)	Netherlands	Prospective birth cohort study	2764 included in the KOALA birth cohort	Organic vs conventional foods from week 34 in pregnancy till child age 2.	Eczema and/or wheeze occurrence.	Use of organic dairy products gave OR 0.64 (0.44,0.93) for eczema at 2 years. No effect on wheeze or atopic sensation.
Smit <i>et al.</i> (2007)	Netherlands	Cross-sectional	593 organic and 1205 conventional farmers.	Farming practice	Hay fever and asthma like symptoms.	Childhood on farms and current livestock farming protected against allergic disorders independent of farming practice.
Abell, Ernst and Bonde (1994)	Denmark	Cross-sectional	30 men involved with organic farming, 73 controls	Members of organic farming association vs. controls.	Sperm quality	Higher sperm concentration among organic farmers.
Jensen <i>et al.</i> (1996)	Denmark	Cross-sectional	55 high-consumers of organic foods, 141 controls	Organic food consumers	Sperm quality	43% higher sperm concentration among organic food consumers.
Juhler <i>et al.</i> (1999)	Denmark	Cross-sectional	85 organic and 171 conventional farmers	Pesticide exposures possible impact on sperm quality.	Semen quality according to intake of organic fruit and vegetables.	The group with no organic food intake had a lower concentration of morphologically normal spermatozoa.
Larsen <i>et al.</i> (1999)	Denmark	Cross-sectional	85 organic and 171 conventional farmers	Pesticide exposures possible impact on sperm quality.	Sperm quality between organic and conventional farmers.	No difference in semen quality.
Christensen <i>et al.</i> (2013)	Denmark	Case-control	306 cases, 306 controls	Use of organic diet in pregnancy	Hypospadias	No differences assoc. with organic food consumption.
De Lorenzo <i>et al.</i> (2010)	Italy	Intervention study	130 men	14 days with Mediterranean diet organic (T2) and conventional (T1).	BMI, Dual-X absorptiometry, blood parameters.	Significant reduction in risk factors for CVD during the T2 period.

### 3.1.1 IMMUNE-ASSOCIATED OUTCOMES

In a cross-sectional study from Sweden, Alm *et al.* (1999) investigated 295 children aged 5-13 years from anthroposophic families attending Steiner schools. The focus was on prevalence of atopic diseases, and they were compared with 380 children attending state schools. There were marked differences between the two groups of children concerning use of antibiotics, MMR vaccination (measles, mumps and rubella), history of measles and consumption of biodynamic food and fermented vegetables. History of atopy or symptoms consistent with atopy was found in 25% of children attending conventional schools compared to 13% from Steiner schools and (OR 0.62 (95% CI 0.43,0.91) for the Steiner school children and adjusted for MMR vaccination, other vaccinations, antibiotics not more than twice and not before two years of age, antipyretics, consumption of fermented vegetables and mainly biodynamic produced foods. When organic food alone was computed against atopy/no atopy there was a significant result towards organic food and reduced risk of atopy (OR 0.63 (95% CI 0.42,0.94). In this study an anthroposophic lifestyle, which includes much more than organic food, indicate a protection against atopic diseases and atopy. The authors concluded that lifestyle factors related to the anthroposophic way of living including organic/biodynamic food appear to reduce the risk of atopic disease in children.

In a prospective epidemiological birth cohort study in the Netherlands, Kummeling *et al.* (2008) investigated whether early life organic food consumption was associated with developing atopic symptoms during the first two years of life. A birth cohort (n=2834) was followed prospectively with detailed questionnaires at 3, 7, 12 and 24 months. The mother's dietary intake from gestational week 34 until delivery and the food intake of the children the first 24 months were registered. Eczema and/or wheeze occurrence were classified by questionnaire, sensitisation by total and specific IgE. A trend for decreased risk for eczema in the organic consumers was seen, but it was not statistically significant. A significant reduced risk of eczema was demonstrated in the group consuming strictly organic dairy products (OR 0.64 (95% CI 0.44, 0.93). The analyses was adjusted for sex, maternal education, body mass index (BMI), parental and sibling history of allergy, number of older siblings, breastfeeding, day-care attendance, pets, exposure to tobacco smoking, vaccination, antibiotic exposure and vegetarian diet. The authors suggested that pregnant and lactating mothers' consumption of organic dairy products within the context of an organic diet is protective of eczema in young children.

Smit *et al.* (2007) performed a questionnaire survey in adult conventional (n=1205) and organic farmers (n=593) from the Dutch part of the European Community Respiratory Health Survey to compare the prevalence of respiratory symptoms and furthermore compare the farming with a non-farming population. Farmers reported significantly less frequent asthma and asthma-like symptoms than non-farmers. Almost all asthma symptoms were less prevalent in organic than in conventional farmers while hay fever was reported more commonly. However this association decreased when adjusting for age, smoking habits, sex, current farm type, childhood farming environment and disinfectant use in a multiple regression model. The study also showed that keeping livestock and growing up on a farm were associated with a two times lower prevalence of hay fever. In conclusion, this study showed that a farm childhood in combination with current livestock farming protected against allergic diseases. This effect was found for both organic and conventional farmers.

### 3.1.2 SEMEN QUALITY

Abell *et al.* (1994) performed a cross-sectional study analysing the semen of 30 members of the Danish Organic Farmers' Association and in 73 different blue-collar workers (19 were

flexoprinters, 22 were electricians and 32 were metal workers), all between 18 to 50 years of age. According to a self-completed questionnaire the average intake of organic dairy products was at least 50% of the total intake of dairy products the past year among 28 of the members. The members of the Organic Farmers Association had significantly higher sperm density compared with the three other groups of workers. The semen volume was; however, lower in the members of the Organic Farmers Association although they had a significantly lower continence period. In the members of the Organic Farmers Association there was a significant correlation between continence period and semen volume, but not sperm density. The total sperm count (density \* volume) was not higher in the members of the Organic Farmers Association than in the other groups. The authors conclude that the higher sperm density among the members of the Organic Farmers Association in spite of shorter continence period was unexpected.

Jensen *et al.* (1996) studied semen quality among 55 members from two different organisations for organic farming in a cross-sectional study in Denmark. All had at least 25% of the diet from organic products and were between 20 to 45 years of age. As controls, 141 men 23-43 years of age, working in an airline company were included. A multiple linear regression of log transformed semen parameters controlling for age, continence period and season showed that the organic food users had 43% higher sperm concentration compared to the controls. No other sperm parameter was different between the two groups. The authors reflect on selection bias since in the organic group members only wanted to participate if they knew they were fertile, while in the control group fertility problems might have triggered participation. The prevalence of genital disorders among the organic food users was lower compared to the control group while reported cryptorchidism was higher.

In another Danish study Juhler *et al.* (1999) analysed the exposure to pesticides in the same study as Larsen *et al.* (1999) (see below), where a food frequency questionnaire (FFQ) was answered in addition to semen collection. The study included 171 conventional farmers, mean age 38 years, and 85 organic farmers, mean age 40 years, who delivered one sample of semen just before the spraying season in 1995/1996. The farmers were divided into three groups where organic food intake represented 0%, 1-49% and 50-100% of the fruit and vegetable intake. Those with a high intake of organic fruit and vegetables also had a high intake of other organic foods. Calculations of exposure to 40 different pesticides were performed with use of the FFQ data, standard portion sizes and concentration of the pesticides collected from the National Danish Food Monitoring Program. All participating farmers were exposed to the pesticides at 1% of acceptable daily intake (ADI) or below the ADI, except for three pesticides where the exposure varied between 1.1% and 2.2% of ADI. The farmers with high organic food intake were significantly less exposed to pesticides compared with those with no intake of organic food. The median sperm count did not vary between the three groups, but the farmers with no intake of organic food had lower concentration of morphologically normal spermatozoa. However, in relation to 14 other semen parameters no significant differences were found. Five significant correlations were found out of 160 analyses with regard to pesticides exposure and different semen parameters. All five significant correlations showed a lower percentage of dead spermatozoa with higher intake of organic foods. The analyses were adjusted for age, semen spillage, sexual abstinence, fever previous three months, smoking, alcohol intake and self-reported reproductive disease. The authors conclude that exposure to the 40 different pesticides analysed did not increase the risk of impaired semen quality.

In the same cross-sectional study Larsen *et al.* (1999), aimed to confirm or refute the hypothesis that organic farmers have higher sperm concentration than conventional farmers. The median sperm concentration was significantly higher in the organic farmers compared to

the conventional farmers, but after adjusting for age, semen spillage, sexual abstinence, fever previous three months, smoking, alcohol intake and self-reported reproductive disease the difference was no longer significant. The conventional farmers had significantly lower count of normal spermatozoa also after adjusting for confounding in one sample of the study, but this difference was not confirmed in a second sample. No significant differences were seen for total sperm count; proportion of non-vital spermatozoa, sperm chromatin structure or motility variables, and the authors concluded that there was no significant difference between conventional and organic farmers with regard to sperm quality.

### 3.1.3 HYPOSPADIA

Christensen *et al.* (2013) performed a case control study in Denmark including mothers of 306 boys operated on for hypospadias and 306 control children matched for geography and child birth year. Through a telephone interview, the intake of fruit, vegetables, milk, dairy products, eggs and meat during pregnancy was recorded and use of organic or conventional products was registered. Intake frequency was categorised as often/sometimes and rarely/never. The telephone interview was carried out in 2005 and 2006 and the hypospadias cases were operated on between 2003 and 2005, meaning that some of the participants were interviewed up to three years after pregnancy. Logistic regression models were constructed for dietary variables and adjusted for maternal age, BMI, and alcohol consumption, but was not adjusted for birth weight. Overall, an organic food choice during pregnancy was not associated with hypospadias in the offspring. However, often consumption of high fat dairy products while rarely or never choosing the organic alternative was associated with increased odds for hypospadias (adjusted OR 2.18, 95% CI 1.09-4.35). The authors refer to exposure of pesticides as a possible causation since some human studies have indicated a possible relationship between pesticides and hypospadias.

### 3.1.4 RISK FACTORS FOR CARDIOVASCULAR DISEASE

De Lorenzo *et al.* (2010) studied the effects of an Italian Mediterranean diet, consisting of organic versus conventional foods in a clinical study. The outcome measures were body composition, and biochemical parameters. The study included 150 healthy Caucasian Italian men (mean age 45 years) and 50 male patients with chronic kidney disease (mean age 46). The aim was to investigate if organic food could decrease cardiovascular disease (CVD) risk factor and the progression of renal diseases. The total number that completed the study was 130 (65%), but information about how many in each group was not available. At baseline, and after 14 days on a conventional Italian Mediterranean diet and after 14 days with organic Italian Mediterranean diet, measurements of BMI, body composition, by Dual-X absorptiometry scanner, the total plasma homocystein (p-tHcy), serum phosphorus and glucose concentrations, lipid profile, and microalbuminuria were performed. The results showed a significant reduction of p-tHcy and phosphorous concentrations during the Italian Mediterranean diet with organic food compared to the Italian Mediterranean diet with conventional food. Body composition analysis showed significant decrease in fat mass after the organic diet, expressed as kilograms and as percentage ( $p < 0.001$ ). Improvement of lean body mass was observed in the patients with chronic kidney disease ( $p = 0.004$ ). The authors concluded that intake of organic Italian Mediterranean diet reduced p-tHcy and microalbuminuria levels in healthy individuals and in patients with chronic kidney disease. However, drop-outs were not reported which might have biased the results. This was the only study found looking at homocysteine and phosphorous concentrations and body composition

parameters with regard to intake of organically produced food, and this paper will not be discussed further.

### 3.1.5 DISCUSSION AND SUMMARY OF THE MAIN FINDINGS, STUDIES IN HUMANS

It is reported that most consumers using organic produced food do this of reasons built on the belief that organic foods are naturally produced, safe and healthy (Hjelmar 2011, Torjusen *et al.* 2001, van de Vijver and van Vliet 2012). The scientific basis for possible human health effects of the consumption of organically produced food is still scarce, and in our literature search only nine studies related to clinical health outcomes were found.

Lifestyle seems to have impact on the development of atopic diseases, and a positive effect of growing up on a farm on atopic disease has been documented before (Riedler *et al.* 2000, von Mutius and Radon 2008, von Mutius and Vercelli 2010). The same finding was reported by Smit *et al.* (2007) and she found lower frequency of atopic disease in organic farmers as well as among conventional farmers, and especially among those growing up with animals. The Alm *et al.* study (1999) showed a positive effect of anthroposophic lifestyle on the frequency of atopic disease and Kummeling *et al.* (2008) demonstrated reduced risk of eczema among children who at the age of two had only been given organic food and were born to mothers who during pregnancy and breastfeeding consumed organic dairy products. The biological plausibility for an impact of organic food on atopic disease and especially eczema is unclear. A higher content of n-3 fatty acids and conjugated linoleic acid in organic than in conventional dairy products was demonstrated (Rist *et al.*, 2007 and Mueller *et al.*, 2010), and it is hypothesised that anti-inflammatory properties of these fatty acids could be protective. Furthermore, it is hypothesised that organic milk contains a higher amount of non-infectious microbial components which might have an effect on gut immunity (Kummeling *et al.* 2008, Suhren and Heeschen 1996).

Health effects of an anthroposophical lifestyle, in which organic/biodynamically grown food is one of several characteristic features has been discussed in several papers. This aspect may be important and can suggest clues to possible health benefits of organic food consumption (and related practices), and it will be interesting to see if future studies may further disclose more details about the specific practices which contributes to the reported findings. If more specific beneficial practices are identified, they may be adopted also in other lifestyle settings and have broader relevance for public health.

Higher sperm concentration in semen among members of the Danish Organic Farmers Association was first seen by coincidence in a study where the authors examined fertility among green-house workers. The first study designed to reevaluate the finding was performed by Abell *et al.*, (1994) followed by Jensen *et al.*, (1996). Both studies indicated a higher sperm concentration in organic farmers compared with conventional farmers or in blue collar workers. Both studies have been criticised for selection bias and for missing statistical adjustments of confounders. Another aspect mentioned was that the collection of semen was performed during different time periods of the year and seasonal variations may have impact on semen quality. In a more comprehensive study no difference was found between organic and conventional farmers with regard to sperm concentration when confounders were adjusted for (Larsen *et al.* 1999 and Juhler *et al.* 1999). In these studies exposure to pesticides from foods could not be separated from the occupational exposure which further complicates the picture and no conclusion was drawn regarding consumption of organically produced foods and semen quality. The suggested mechanism for an impact on sperm count and sperm quality has been pesticide exposure and it has been shown that high exposure to pesticides reduces sperm count and quality (Hossain *et al.* 2010). In the study by Juhler *et al.* (1999) it

was shown that the conventional farmers were exposed to more pesticides than the organic farmers. However, the exposure was only between 1-2.2% of ADI for some of the calculated pesticides among conventional farmers, and in a recently published study from Norway no impact on sperm quality of pesticide exposure could be measured (Haugen *et al.* 2011). Hypospadias is another health outcome that has been connected with pesticide exposure and in a recent Greek study mothers giving birth to a boy with hypospadias were more highly exposed to pesticides than mother with boys born without hypospadias (Michalakis *et al.* 2013). Pesticide exposure through food might also explain the findings in the Danish study (Christensen *et al.*, 2013). A general healthier lifestyle related to choosing organic alternatives was also suggested as an explanation of the findings.

To summarise, two studies indicated a positive effect of lifestyle factors including organic food use and atopic diseases in children. However, the importance of organic food use could not be disentangled from other lifestyle factors. A lower exposure to pesticides with organically compared to conventionally produced food was discussed with regard to sperm quality and hypospadias, but no conclusion could be drawn. The shortage of clinical as well as epidemiological well designed studies in humans implicate that no conclusions could be drawn with respect to health outcomes.

### 3.2 ANIMAL MODEL STUDIES

Nine studies from the literature search described animal models used for comparing organically grown crops to conventionally grown. The study design varied, as some studies took seasonal and yearly difference into account and others not. Furthermore some studies used blinding of the investigators, others not. Animals studied were rats (six studies), mice, chicken and *Drosophila Melanogaster* (fruit flies) (one study each). Various health parameters were tested, but all studies included some immune parameters. One of the studies (Huber *et al.* 2010) aimed at identifying markers of health which may be used for future human intervention studies comparing organically and conventionally produced food. Another study (Lauridsen *et al.*, 2008) aimed at defining which measurable aspects of health might be affected by differences in production methods. Table 3.2 gives an overview of all the animal model studies included in this opinion.

**Table 3.2: Overview of the animal model studies included in this opinion.**

Authors	Country	Study design	Type of animal	Study characteristic	Main endpoint	Main findings
Chhabra <i>et al.</i> (2013)	USA	10 day feeding of organic vs conv. foods.	Fruit flies ( <i>Drosophila Melanogaster</i> )	Observed for 60 days after the feeding period.	Longevity, fertility and oxidative stress resistance.	Longer life span, higher fertility and higher resistance to oxidative stress when fed organic products.
Jensen <i>et al.</i> (2013)	Denmark	Effect of maternal feed with organic vs conventional crops during pregnancy and lactation on oral tolerance development in offspring.	Rats	Followed for 2 generations. First generation on organic or conv. Feed.	Response in offspring to a novel dietary antigen (ovalbumin).	Cultivation system had no effect on organ weight and immunological biomarkers.
Srednicka-Tober <i>et al.</i> (2013)	Poland/ UK	Feeding with crops from 4 different production systems.	Rats	Fed for 9 weeks on experimental diets. Parents were on same diet.	Body composition, growth, humoral and immune status.	Crop production (fertilisation and crop protection) had significant effects on animal physiology, especially immune status.

Authors	Country	Study design	Type of animal	Study characteristic	Main endpoint	Main findings
Jensen <i>et al.</i> (2012)	Denmark	Feeding 40% organic vs conv. carrots performed 2 years.	Rats	First year experiment 95 days. Second year 63 days.	Biomarkers of health and immunity.	No differences between the feeding groups.
Roselli <i>et al.</i> (2012)	Italy/ Denmark	Feed consisting of 40% organic vs 40% conv. carrots. 2 years crop.	Mice	21 day old mice were fed for 30 days both years.	Biomarkers of health and immunity.	Immune stimulation was found after organic carrot consumption but no difference in inflammatory status.
Skwarlo-Sonta <i>et al.</i> (2011)	Poland/ UK	Feeding organic and conv. crops cultivated in four different production systems. Two consecutive years.	Rats	Experiment for 12 weeks each year.	Growth, hormonal conc. immunological parameters.	The production method affected the feed composition, the growth of the rats, hormonal balance and immune status.
Huber <i>et al.</i> (2010)	Netherland	Organic vs conv. diet fed to 2 generations and in 3 chicken lines.	Chicken	Fed conv. diet until age 11 weeks and then experimental diet for 94 days. Second generation was studied to rule out epigenetic confounders.	Immune parameters (innate and specific). Genomic analyses.	A tendency toward an enhanced immune competence with organic feed. 30 genes were differentially regulated between the feed groups.
Lauridsen <i>et al.</i> (2008)	Denmark	Two generations on 3 different feeding regimen.	Rats	2. generation was tested during week 19 to 44.	Daytime activity, body composition, immunological parameters and fatty acids in adipose tissue, liver and blood.	Rats fed the organic diet had lower daytime activity and lower adipose tissue and had higher IgG concentrations.
Finamore <i>et al.</i> (2004)	Italy	30 days feeding of organic vs conv. wheat to study toxic elements with respect to mycotoxins.	Rats	Two arms with or without adequate protein for 30 days.	Intestinal and splenic lymphocyte proliferative capacity and liver acute-phase reaction.	No difference in proliferative capacity of lymphocytes in well-nourished rats. Lymphocytes from PEM rats fed conv. diet showed lower mitogen stimulation capacity.

### 3.2.1 IMMUNE-ASSOCIATED AND GENERAL HEALTH PARAMETERS

In an animal *in vivo* and *in vitro* study from USA, Chhabra, Kolli and Bauer (2013) studied longevity, fertility and oxidative stress resistance in *Drosophila Melanogaster* fed conventionally or organically grown potatoes, raisins, bananas and soybean during 10 days, and observed up to 60 days. Organic potatoes, raisins and soy led to significantly extended life span compared to conventional, while for bananas the life span was equal. Flies fed organic potato or banana, but not raisins, survived the oxidative stress treatment longer than the flies on conventionally produced feed. Physical activity of the flies was measured over a 48 hour period. It turned out that flies fed organically produced raisins and banana had higher overall activity than the same food produced conventionally. Flies fed extracts of any organic produce had significantly more eggs per day than flies fed on conventional produce. The data suggest that organic food provide improved health outcomes in *Drosophila Melanogaster*. The exact molecular mechanisms of the observed health effects are unclear. Altered insulin-signaling, altered redox balance or xenohormesis may all play a role.

In a blinded animal feeding experiment in two generations of female Wistar Hannover GALAS rats (n=54 first generation, n=29 mothers, n=134 pups), Jensen *et al.* (2013) studied the effect of maternal intake of conventionally or organically produced feed on oral tolerance

development in the offspring. The diets were composed of crops from a long-term field study including two cultivation systems x 2 years x 2 locations = 8 diets. The crops used were potatoes, winter wheat, spring barley and fava beans grown with organic cultivation system with green manure and without pesticides or conventional with inorganic fertilisers with pesticides. No differences in immune parameters or induction of oral tolerance between offspring rats of mothers fed organically or conventionally grown plant diet were observed. The conclusion was that the data indicate an influence of maternal diet during pregnancy and lactation on tolerance development in the offspring, but no differences were observed between organically or conventionally grown diets.

In an animal feeding study from Poland, Srednicka-Tober *et al.* (2013) studied rats fed crops from four different crop management regimes: 1) organic fertilisation and crop protection, 2) organic fertilisation and conventional crop protection, 3) conventional fertilisation and organic crop protection and 4) conventional fertilisation and crop protection. The parents of the rats studied were fed on crops from the same regimes as the offspring. The aim of the study was to quantify the effects of different cultivation regimes on composition of rat feeds, body composition, growth, hormonal and immune status parameters.

There were no detectable pesticide residues except for the growth regulator chlormequat which was detected in wheat grain grown under conventional crop protection. There were lower protein and cadmium concentrations in organically fertilised crops, higher concentration of polyphenols.

Rat growth was similar for all types of feed and final body weight was not affected by treatment. Organic fertilisation resulted in higher body weight at weaning, but lower total weight gain. Body fat content was not affected by type of feed.

Hormonal balance was affected by the type of feed; Leptin and Insuline-like growth factor-1 (IGF-1) was higher in rats fed crops organically fertilised and with organic crop protection, corticosterone level was higher and testosterone lower in rats fed organically fertilised crops.

Several immune parameters were affected by the type of feed: Lipopolysaccharid stimulated lymphocyte proliferation was low with no stimulation of cells from rats on organically fertilised crops. Feeds from organically fertilisation and organic crop protection resulted in an increase in spontaneous proliferation, while organic fertilisation and conventional crop protection led to a decrease in Con-A stimulated proliferation. No significant effect on IgA or C-reactive protein (CRP) by crop management regimes.

The authors conclude that different crop production practices which result in modulation of the composition of feed, may have a significant effect on rat physiology, especially immune status.

Jensen *et al.* (2012) investigated in an *in vivo* and *in vitro* animal study the impact of consumption of carrots as whole food, grown under organic or conventional cultivation system during two consecutive years in Denmark could have on health parameters in female GK/MolTac (n=65/year) rats. The conventionally produced carrots were grown using inorganic fertiliser and pesticides. Three different organic cultivation systems were studied: minimalistic organic cultivation system following organic practices, with the maximum allowed manure input (O1), organic cultivation with intensive use of cover crops (O2) and (very organic) cultivation using green manure, cover crops and intercrops (O3). The cultivation system and year did not show any impact on biomarkers of health and immunity. The conclusion was that the study showed little impact of cultivation system and year on carrot quality and little (if any) effect of cultivation system on biomarkers of health and immunity.

Roselli *et al.* (2012) investigated the impact of organic and conventionally grown carrots on intestinal and peripheral immunity in mice in an *in vitro* study from Italy. The study comprised two production years, and three varieties of organic production grown in two different geographic locations (Denmark and Italy). The carrots were grown in four different cropping systems for vegetable production, one conventional and three organic (O1-3). The O1 was similar to the conventional but organic fertiliser and no pesticides, O2 and O3 had increasing content of fertility-building crops. The study indicated an immune stimulation by the organically grown carrots, especially from the two “more organic” produce. The results from measuring pro- and anti-inflammatory cytokines in serum showed no significant differences between conventional and organic groups of the three varieties. The authors suggest that although the study was not designed to study safety, the fact that organic feed did not induce an inflammatory response, could be an indication that organically produced carrots did not introduce toxic compounds in mice. In conclusion it is stated that the study indicate an immune stimulation by organic carrots on intestinal and peripheral immunity, including an expansion of regulatory T lymphocytes. The study reinforces the concept that organic products are as safe as conventionally grown.

Skwarlo-Sonta *et al.* (2011) performed a dietary intervention study in male Wistar rats (n=96/year) fed crops grown under conventional or organic conditions during two consecutive growing seasons. The crops were grown in the UK and transported to Poland where the feeds were prepared and the experiments carried out. The total feeding period on experimental diets was 12 weeks. The crops were produced under four different production systems: 1) organic 2) organic crop protection + conventional fertilisation 3) conventional crop protection + organic fertilisation 4) conventional. In the first year no differences in rat growth were found, in the second year the growth rates of both generations rats were higher in rats fed crops grown conventionally. No differences between organic or conventional systems in body composition or hematological parameters. Plasma antioxidant capacity measured as trolox equivalent antioxidant capacity (plasma TEAC) was higher in rats fed crops produced under organic crop protection regimes. Differences in IgA and IgG in serum was detected with higher levels of IgA in rats fed on conventionally produced crops while differences in IgG levels were only seen between years and not between production methods. Organic crop protection and organic fertilisation methods resulted in higher levels of proliferation after ConA stimulation of lymphocytes. In conclusion the results of the study show that production method affected the composition of the rat feeds, the growth of the rats, their hormonal balances, body composition, and the immune status. However, the differences in feed composition could not be directly linked to the observed physiological differences.

In a blinded animal feeding experiment from the Netherlands in two generations of chickens (n=72 first generation, n= 150 second generation), Huber *et al.* (2010) aimed at identifying biomarkers for future human intervention studies of the effect of organic feed on health in a chicken model, focusing on immune parameters (innate and specific). The study was carried out in three chicken lines, a high immune responder line (H), a mixed conventional line (C) and a low immune responder line (L). Furthermore two generations were studied, focusing mainly on the second generation in order to rule out epigenetic confounders. All animals were fed a commercially obtained chicken diet until 11 weeks of age, from 11 weeks – 94 days the animals received the experimental diets. Body weight differed significantly in the C-line animals, the conventionally fed being heavier during the whole period. L-line animals in the first generation on organic feed displayed a significant increase in Newcastle Disease vaccine-specific antibody titers. In the second generation the organically fed appeared to have a stronger immune competence in the innate immune system reflected by an enhanced response of monocytes after Keyhole Limpet Haemocyanine (KLH) challenge. The challenge activated

the specific immune system stronger, but no difference between the feed groups was seen. The different immune parameters indicate an immune modulation by the feeds and tendencies towards an enhanced immune responsiveness or immune competence of the animals on organic feed. Intermediates of the lipid metabolism showed differences in the organic group after KLH challenge, in accordance with the immunological findings. The authors speculate that the differences found may partly be explained by more immune-stimulating gram-negative bacteria in the organic feed, but the differences between feeds were limited and the quantitative addition of bacteria is small compared to the bacterial load present in the gut. Genomic analyses displayed thirty genes differentially regulated between the feed groups independent of their genetic background. Of these, seven genes were involved in cholesterol biosynthesis and up-regulated in jejunum from the organically fed animals. Other genes found were involved in immunological processes. The authors concluded that diets from different cultivation systems can induce physiological changes in two generations of chickens. The underlying mechanisms are unclear.

Lauridsen *et al.* (2008) performed a rat feeding study in Denmark using diets from crops cultivated with three varieties of nutrients and pesticide use. Their aim was to identify health-related biomarkers affected by different growth conditions of plant foods. The production methods used in the study were not the same as actual commercial systems. One diet was produced using similar or lower levels of organic fertiliser input through plant residues and animal manure than standard organic practice with no pesticides, and one diet corresponded to standard conventional practice. The third method was organic in fertiliser input, but used pesticides. Although the dietary treatments were similar in terms of nutritional quality, there were differences in alpha-tocopherol and IgG, daytime activity, volume of adipose tissue, liver metabolic function and liver peroxidation. The rats fed an organic diet had a generally lower activity during the day with no differences during night. Rats being night-active animals usually rest during daytime and the result indicate a more uninterrupted sleep in organic fed rats which is in accordance with the lower content of adipose tissue in these rats. The higher IgG concentrations in rats fed organic and organic with pesticides compared to conventional diet is in accordance with studies claiming that immune function could be affected by differences in cultivation systems. The authors concluded that the study has identified biomarkers which should be taken into account in further well designed studies in this field.

Finamore *et al.* (2004) studied male Sprague-Dawley rats in a 30 days feeding period of organic vs conventionally grown cereals. The aim of the study was to propose a novel approach to evaluate the potential health risk induced by long-term consumption of organic versus conventional cereals, that is, the assay of sensitive markers of cell function in vulnerable conditions, in which cell defense can be less efficient. The rats were randomly assigned to two groups fed conventional or organic wheat. Each group was divided in two subgroups of well-nourished or protein-energy-malnourished (PEM) rats. The well-nourished rats were fed a complete balanced diet containing 24% casein and 8% wheat protein. The PEM subgroups were fed a casein free diet where 8% wheat protein represented the only protein source. The lymphocytes were cultured in medium containing fetal calf serum as a standard serum and also in medium containing the corresponding rat serum to mimic the *in vivo* conditions. The proliferative response of lymphocytes collected at day 30 after sacrificing the animals was measured in the four subgroups. No differences in proliferative capacity of lymphocytes (peripheral and intestinal sites) were seen in well-nourished rats. Organic wheat did not affect the proliferative capacity of lymphocytes in well-nourished or PEM rats when cultured in medium containing fetal calf serum. Lymphocytes from PEM rats cultured in rat serum, however, showed lower mitogen stimulation capacity in the

conventional fed than in the organic fed. The authors suggest that the latter observation indicates the presence of contaminants in conventionally grown and not in organic wheat. The overall conclusion was that this study demonstrates no higher risk of organic compared to conventional wheat that could affect fundamental cell functions. Furthermore, the study introduces the concept that it is advisable to use functional assays as well as chemical in the evaluation of food health risk.

### 3.2.2 DISCUSSION AND SUMMARY OF THE MAIN FINDINGS IN ANIMAL MODEL STUDIES

The animal model studies included in this report were intended to be models for human health and thus either describe physiological processes which may directly pertain to humans or search for biomarkers of health which may in future be studied in humans. The studies differed in design, animals and crops for feed and it was thus difficult to compare and evaluate the results. There are, however, some general traits which can be considered.

The immune parameters investigated in animal studies were immunoglobulin concentrations and spontaneous proliferative capacity of peripheral and gut/spleen lymphocytes with the different feeds or after stimulation with mitogens. Roselli *et al.* (2012) found a stronger immune stimulation by organic carrots, a result supported by Finamore *et al.* (2004) in well-nourished rats fed organic cereals, but not in protein-energy malnourished rats. Skwarlo-Sonta's study in rats showed that rats fed organic crops had a higher proliferative response after Concanavalin-A stimulation (Skwarlo-Sonta *et al.*, 2011). The latter finding is different than the findings in Srednicka-Tober *et al.* (2013) who demonstrated that organic fertilisation did not influence Con-A - stimulated proliferation under organic crop protection, but resulted in lower proliferation under conventional crop protection. On the other hand Srednicka-Tober *et al.* also demonstrated a significantly higher spontaneous lymphocyte proliferation with organic fertilisation and crop protection regimes, but no effect of organic fertilisation under conventional crop protection regimes. The differences found in organic vs conventional feed on immune parameters as lymphocyte proliferation, spontaneous or mitogen stimulated, may be explained by the presence of other unidentified immune-modulating properties in feeds, as also suggested by Finamore *et al.* (2004). Furthermore, yearly differences had an even more marked effect on mitogen stimulation, both by Concanavalin-A and Lipopolysaccharid. Thus, it is important to study yearly variations as well as cultivation method. Huber demonstrated a higher antibody titer after Newcastle Disease Vaccination in a low immune response strain of chicken on organic feed, a result which is in accordance with the finding of higher IgG concentrations in organically fed rats (Lauridsen *et al.*, 2008). However, two studies by Jensen *et al.* (2013 and 2012) did not find any differences in immunological parameters between organic and conventional feed in rats.

A reduction in adipose tissue was demonstrated by Lauridsen *et al.* (2008) in rats fed organically and Huber *et al.* (2010) (chicken). This is in contrast to Skwarlo-Sonta *et al.* (2011) who demonstrated lower body fat in rats fed on diets based on compound feeds made from conventionally fertilised crops and to the study by Srednicka-Tober *et al.* (2013) where no effect of diet on body fat was recorded. Huber *et al.* also showed an improved post challenge catch-up growth for chicken fed organic products, in contrast to Srednicka-Tober *et al.* who found a higher body weight at weaning in rats fed crops under organic fertilisation, but a lower total weight gain. These results were in contrast to the studies by Jensen *et al.* (2013 and 2012) where all rats were clinically healthy with little or no impact on biomarkers of health by cultivation methods for the feeds. Likewise the study by Finamore *et al.* (2004) did not detect differences in general health parameters. Interestingly, Chhabra *et al.* (2013) studied the activity in fruit flies (*Drosophila Melanogaster*) on organic or conventional feed

and demonstrated a higher overall activity in flies fed organic raisins and banana food. On the other hand, Lauridsen *et al.* (2008) found that rats, being night active animals, had a lower daytime activity when fed organic feed, whereas the nightly activities of the rats were similar to those on conventional feed. The authors interpreted the finding with organically fed rats having more uninterrupted rest during the daytime which may explain their lower content of adipose tissue. This is in line with human studies showing a significantly increased weight gain in people having less than seven hours sleep per night (Patel *et al.* 2006).

Although differences have been shown in plasma concentration of alfa- and gamma tocopherol between rats fed conventional and organic diet, the long-term status of vitamin E as assessed by measurements in adipose tissue showed no effect of dietary treatment (Lauridsen *et al.*, 2008), indicating adequate amounts to meet physiological needs in all diets, and thus probably no direct implication for health. The results are in accordance with those of Jensen *et al.* (2013 and 2012)

There have been concerns that the reduced use of pesticides and fungicides in organic farming may lead to a greater contamination by toxins in organic crop, while others tend to claim the opposite. Finamore *et al.* (2004) found that the proliferative capacity of rats fed organically and conventionally grown feeds were similar, and furthermore that organic wheat did not affect the proliferative capacity of lymphocytes from protein-energy-malnourished rats with a less effective immune response. Finally, when lymphocytes from protein-energy-malnourished rats were cultured in rat serum to mimic *in vivo* situation, the capacity to respond to mitogen stimulation was lower in conventional protein-energy-malnourished rats than in organic protein-energy-malnourished rats. The latter finding indicates more contaminants present in conventionally than organically fed protein-energy-malnourished rats. Roselli *et al.* (2012) performed a cytokine secretion study which showed that organic carrots did not induce an inflammatory status in mice which could be an indirect indication of safety. However, the study was not designed for investigating safety.

To summarise, in some well performed animal studies, a positive effect on animal physiology which may have an impact on animal health like immune parameters, hormonal balance and activity level was shown. Not all studies substantiate these findings. Explanation for the discrepancies is to be found in the differences in study design which include animal breed, plant food used, growing seasons and geographical location.

### 3.3 BIOMARKER STUDIES

Major findings from thirteen papers are described in this section. Most studies on the blood biomarkers for antioxidant status were based on small number of subjects. The study designs and biomarkers measured in these investigations varied and the findings based on these studies may be used with caution. Table 3.3 gives an overview of biomarker studies included in this opinion.

**Table 3.3: Overview of biomarker studies included in this opinion.**

Authors	Country	Study design	Study characteristic	Main endpoint	Main findings
Soltoft <i>et al.</i> (2011)	Denmark	Crossover study to investigate the impact of 2 organic diets vs conv. diets.	3 x 12 days diet periods with wash out of 2 weeks. 18 subjects participated.	Carotenoid concentration in plasma.	Plasma concentration increased with all three diets, but no significant difference between organic and conv. diets.

Authors	Country	Study design	Study characteristic	Main endpoint	Main findings
Burns-Whitmore <i>et al.</i> (2010)	USA	3-armed single-blind randomised crossover study to study lutein bioavailability from n-3 enriched and organic eggs.	20 healthy lacto-ovo-vegetarians eating either n-3 enriched eggs or organic eggs or no eggs 6 days a week for 8 weeks.	Serum lutein concentration after each period.	Serum lutein was significantly higher after the egg eating weeks ( $p < 0.009$ ), but no difference between egg treatment groups.
Stracke <i>et al.</i> (2009)	Germany	A double-blind randomised intervention study of carrots.	36 volunteers consumed 200 g carrots per day for two weeks.	Bioavailability antioxidant capacity and endogenous DNA strand breaks.	No difference between groups with regard to total carotenoid concentration, antioxidant capacity or endogenous DNA strand breaks.
Briviba <i>et al.</i> (2007)	Switzerland	A double blind randomised, cross-over study with apples.	Six non-smoking healthy men consumed 1000 g organically or conventionally cultivated apples for 2 days with 1 week washout period in between.	Antioxidant capacity, LDL and endogenous DNA strand breaks.	No differences between organic or conventionally grown apples were seen for antioxidant capacity, or LDL. Similar antigenotoxic potential of both organic and conv. grown apples.
Di Renzo <i>et al.</i> (2007)	Italy	A clinical study to evaluate antioxidant variables.	10 healthy men first consumed a conv. diet for 14 days and then a similar but organic for 14 days.	Antioxidant capacity	A significant increase in antioxidant capacity after the 14 days with organic diet ( $p < 0.005$ ).
Akçay <i>et al.</i> (2003)	Turkey	A clinical study to investigate consumption of red wine and antioxidant capacity.	Six healthy men consumed 200 ml and two healthy women took 100 ml red organic wine in 15 minutes and blood samples were drawn for 6 hours.	Total antioxidant activity and erythrocyte thiobarbituric acid reactive substance.	The two wines did not differ in the ability to inhibit the oxidation of LDL.
Caris-Veyrat <i>et al.</i> (2004)	France	A randomised clinical trial to evaluate antioxidant capacity in tomatoes.	10 healthy females ate 100 g/per day of tomato puree for 3 weeks.	Antioxidant capacity	No difference was found between the two purees with regard to their ability to affect plasma levels of the two major antioxidants, vitamin C and lycopene.
Grinder-Pedersen <i>et al.</i> (2003)	Denmark	Double-blind crossover intervention study and effect on markers of five flavonoids and of oxidative defense.	6 men and 10 women, all non-smokers, volunteered to eat an identical organic and conv. diets for 22 days.	Antioxidant capacity and urinary excretion of five flavonoids.	Excretion of quercetin and kaempferol was higher in the organic diet group ( $p < 0.05$ ). The excretions of flavonoids were similar and most markers of antioxidative defense. Decreased total plasma antioxidant capacity compared to baseline with organic diet ( $p < 0.05$ ).
Rist <i>et al.</i> (2007)	Netherland	Cross-sectional study of breastmilk from 312 women. The KOALA study.	186 mothers used conv. diet, the other <50% or 50-90% and some used a mixture of all organic food.	Breastmilk 1 month after birth. Fatty acid composition.	Women consuming an organic diet had significantly higher rumenic and trans-vaccenic acid concentration in breastmilk, but lower alpha-linolenic acid.
Mueller <i>et al.</i> (2010)	Germany	Cross-sectional study of breastmilk among organic diet users. The KOALA study.	310 mothers with different coherence to organic diet.	Fatty acid composition in breastmilk 1 month postpartum.	Higher concentrations of several trans fatty acids with organic diet, reflecting the fatty acid intake of the mothers.
Mark <i>et al.</i> (2013)	Denmark	Two double blind crossover trials analysing absorption of zinc and copper from organic diet.	17 healthy non-smokers. 3 dietary periods of 12 days. On day 8 isotope labelled breakfast, lunch and dinner were given.	Tracing the isotope Cu and Zn during day 9-12.	No difference in intake or absorption of Cu and Zn between organic and conv. Diet.
Lu <i>et al.</i> (2006)	USA	Clinical study to study organophosphorous pesticides in children.	23 children were give organic diet for 5 consecutive days.	Urinary monitoring of pesticides.	Organic diet reduced urinary malathion and chlorpyrifos to a non-detectable level in all children and a reduction in all the measured pesticides.

Authors	Country	Study design	Study characteristic	Main endpoint	Main findings
Curl <i>et al.</i> (2003)	USA	A cross-sectional study measuring concentration of 5 pesticides in 46-47 months old.	18 children consumed organic diet and 21 a conventional diet.	24-urinary excretion of 5 pesticides	The children consuming an organic diet had lower concentration of 4 of the pesticides and its metabolites. Residential pesticides had no effect on the results

### 3.3.1 ANTIOXIDANT ACTIVITY

Soltoft *et al.* (2011) studied whether crops grown with conventional or with organic agriculture systems using animal manure (OA) or cover crops (OB) had an impact on the content of carotenoids in carrots. Furthermore, the investigators tested the effect of diet based on conventional, OA and OB foods on the concentration of carotenoids in human plasma. The carrots were cultivated with conventional, OA and OB agriculture systems separately in 2007 and 2008. In the carrots, the content of  $\beta$ -carotene was 75%,  $\alpha$ -carotene 23% and lutein 1.9% respectively. The content of carotenoids in carrots cultivated with the three different agriculture systems were not significantly different. Eighteen men with mean age 25 years and average BMI at 22 kg/m<sup>2</sup> (1<sup>st</sup> year) and 18 men with mean age 26 years and average BMI at 24 kg/m<sup>2</sup> (2<sup>nd</sup> year) participated in double blind, crossover intervention trials. The intervention period for the trials was 12 days and a washout period two weeks. The conventional or organic diets based on carrots, onions, white cabbage, barley, potatoes, tomatoes and wheat had similar concentrations of carotenoids. Two menus and six types of diets were prepared from these food items. Six subjects consumed conventional diet, six subjects consumed diet OA and six subjects consumed diet OB. Fasting blood samples were collected in the morning, on day one and on day 13. The  $\beta$ -carotene concentrations in the plasma samples were significantly higher than the  $\alpha$ -carotenoid and lutein concentrations. The concentration of  $\beta$ -carotene increased in plasma samples after the intervention, both during the first and second year. No significant difference was detected in the concentration of plasma carotenoids when the participants with the three diet groups were compared.

The focus in Burns-Whitmore *et al.* (2010) study from the USA was to evaluate the effect of egg intake on antioxidant levels, knowing that oxidative stress is important in the pathogenesis of age-related macula degeneration (an eye-disorder). In their three-armed randomised study they examined plasma antioxidant levels, using high-performance liquid chromatography, in 20 healthy subjects eating either eggs from conventional fed chicken enriched with n-3 fatty acids or eggs from organically fed chicken, whereas the controls did not eat eggs. The participants ate six eggs per week during eight weeks. Serum lutein concentration increased significantly (about 2.5-fold) after the egg consumption period and no significant difference were found between the groups.

Stracke *et al.* (2009) compared the content of carotenoids in carrots cultivated with conventional or organic agriculture systems. The authors also studied the effect of carrot consumption on antioxidant status, DNA strand breaks, and parameters of the immune system in healthy subjects. The content of carotenoids and the lipophilic antioxidant activity, measured as ( $\mu$ mol trolox equivalents/g fresh weight) were not significantly different in the carrots produced organically and conventionally. Thirty-six non-smoker men, age 19-54 years, without the history of medication, participated in a randomised double-blind study. Two groups (12 subjects per group) consumed meals with 200g blanched carrots cultivated organically or conventionally for two weeks, whereas the control group consumed carrot-restricted meals. The blood samples were collected on day 0, 2, 7 and 14. The concentration

of  $\alpha$ -carotene and  $\beta$ -carotene in plasma increased significantly, however, there was no significant difference in the concentration of  $\alpha$ - and  $\beta$ -carotene between the groups eating carrots. The different diets had no significant effect on antioxidant status; ferric reducing ability of plasma, oxygen radical absorbance capacity, trolox equivalent antioxidative capacity (TEAC). Similarly, no significant difference was detected in DNA strand and the carotenoid concentration in the blood mononuclear cells.

Briviba *et al.* (2007) performed a double-blind randomised, crossover study to investigate whether consumption of organically or conventionally produced apples will affect antioxidant status and DNA damage level in healthy subjects. Conventional and organic apples, Golden Delicious, were cultivated according to the criteria established by the Bio Suisse and Suisse Garantie, Switzerland. Eight organically and eight conventionally cultivated apples had similar concentrations of phenolic compound, chlorogenic acid, and sum of all phenolic compounds. Six non-smoking healthy men, mean age 27 years and average BMI 23 kg/m<sup>2</sup> consumed 1000 g organically or conventionally cultivated apples. The subjects did not consume foods containing high concentration of polyphenols for three days before the intervention. The washout period between the two interventions was one week. Blood was collected before and 1, 2, 3, 4, 5, 6, 9, 12, and 24 hours after the intervention. No differences in antioxidant status in plasma were found after consumption of organic or conventionally grown apples. Similar antigenotoxic potential was observed in both organic and conventionally grown apples.

Di Renzo *et al.* (2007) determined the oxygen radical absorbing capacity (ORAC), antioxidant activity, in organic and conventional fruits and vegetables. Except tomatoes and lettuce, the organically produced garlic, orange, banana, carrot, beans, strawberry, lettuce, lemon, apple, potato, tomato, pears, peas, celery, wine, courgettes (summer squash) and milk showed significantly higher ORAC than the conventional products. The authors further tested whether the consumption of diets based on organic or conventional food products might have an effect on the plasma levels of ORAC. Ten healthy non-smoker men, age 30-65, were given diets based on organic or conventional foods for 14 days. ORAC levels in plasma were elevated in men after the consumption of diets based on organic products. The authors suggested that phenolic compounds in organic foods contribute to increase in antioxidant capacity in plasma. However, this study stands alone and will have to be replicated by another research team to get scientific impact.

Akcay *et al.* (2004) investigated whether the consumption of an organic red wine compared to a non-organic red wine increases antioxidant capacity and has an impact on the oxidation of low-density lipoprotein in healthy individuals. Six non-smoker healthy men and two non-smoker healthy women, age 24-45 years, participated in the study. The men consumed 200 ml and the women consumed 100 ml red organic wine in 15 minutes. Blood samples were collected at the baseline, one and six hours after the wine consumption. After a washout period for six weeks, the intervention was repeated. The total content of phenolic compounds, measured as galic acid equivalents, in organic and non-organic red wine was 18.3 mg/ml and 40.2 mg/ml respectively. The peak plasma concentration of galic acid equivalents was reached one hour after the consumption of organic wine and six hours after the consumption of non-organic wine (both 350mg/L). The consumption of organic wine was associated with a significant increase in erythrocyte superoxide dismutase after one and six hours of the trial and in erythrocyte catalase (eCAT) only six hours after the trial. Whereas, the consumption of non-organic wine was only associated with an increase in erythrocyte superoxide dismutase after six hours. Organically and conventionally produced wines did not differ in the ability to inhibit the oxidation of LDL in the absence or presence of CuSO<sub>4</sub>.

Caris-Veyrat *et al.* (2004) determined antioxidants in tomatoes cultured organically and non-organically in one area in France. The plasma antioxidant concentrations were measured with high-performance liquid chromatography, in humans consuming purees based on the two culture-forms of tomatoes. Ten healthy females ate 100 g/day for three weeks of tomato puree (for lunch or dinner) in a randomised trial. It was reported that albeit higher antioxidant levels were present in the organic tomatoes, it did not result in higher plasma antioxidant levels compared to conventional grown tomatoes.

In a double-blind, crossover study, Grønder-Pedersen *et al.* (2003) compared the effect of diets based on organically or conventionally produced foods on urine flavonoids and plasma markers of antioxidant defence system in healthy subjects. Six non-smoker healthy men and 10 non-smoker, non-pregnant women, age 21-35 years, mean BMI 23.4 kg/m<sup>2</sup>, participated in the study. Their average energy intake was 12±2.7 MJ. The subjects consumed diets based on four menus, which contained breakfast, lunch, dinner and snacks. The menus and food quantities in organic and conventional diets were identical. The vegetables grown organically or conventionally were harvested within the same week. Intervention period lasted for 22 days and the washout period was three weeks. Food analyses were performed on duplicate portions of each menu on day one to day four in the first week of each intervention period. Blood samples were collected before the intervention day -1, the baseline 0, after 22 and 23 days after each intervention. 24-hour urine samples were collected in acid-washed containers on day 0 and on day 22. Among the five flavonoids analysed, the quercetin concentration in the organically produced foods was significantly higher than in the conventionally produced foods. The trolox equivalent antioxidant capacity was significantly higher after the consumption of conventionally produced foods than the organically produced foods. Other plasma markers of antioxidant status did not change significantly. The consumption of organically produced foods caused a higher urinary quercetin and kaempferol excretion than the conventionally produced foods. It was suggested that organically produced foods had greater impact than conventionally produced foods on the concentration of urinary flavonoids and markers of antioxidant system.

### 3.3.2 FATTY ACIDS

Rist *et al.* (2007) investigated whether the inclusion of organic dairy and meat products in maternal diet would alter the concentration of conjugated linoleic acid isomers (CLA) and trans-vaccenic acid (18:1 trans-11) in breastmilk from 312 women of KOALA Birth Control Study (The Netherlands). Trans-vaccenic acid is the main trans fatty acid isomer present in milk fat. Mammals convert it into rumenic acid, a conjugated linoleic acid. Based on a self-administered questionnaire 186 women reported that they consumed conventional diets, 33 women consumed diets which included 50-90% organic foods and 37 women consumed diets with >90% organic dairy and meat products. The latter group consumed more dairy fat than the former groups. The participants provided breastmilk samples one month post-partum for the fatty acid analysis. The concentration of rumenic acid (18:2 cis-9, trans-11) was significantly higher also after adjusting for several variables, in breastmilk collected from the women who consumed >90% organic dairy and meat products than in breastmilk from the women who consumed conventional diet. Similarly, trans-vaccenic acid concentration was higher in the breastmilk collected from the women who consumed organic diet than in the women who consumed conventional diet. The breastmilk from the women consuming the organic diet had lower level of  $\alpha$ -linolenic acid (18:3 n-3) and total mono unsaturated fatty acid than the women on the conventional diet. An increase in intake of organic food was associated with increased trans-vaccenic acid concentration. It has not been established yet

whether increased intake of CLA and trans-vaccenic acid by human newborn has any health benefits, nevertheless, in animal models, CLA and trans-vaccenic acid have shown some anti-inflammatory effects.

Mueller *et al.* (2010) measured the ratio of the trans fatty acid isomers in breastmilk from the women from the same Birth Control Study (KOALA Birth Control Study, the Netherlands). The concentration of total trans fatty acids was not significantly different in the breastmilk collected from the women on conventional diet (n=185) than in the women on 50-90% organic diet (n=33) or women on more than 90% organic diet (n=37). However, the ratio 18:1 trans-9/18:1 trans-11 was lower in breastmilk from the women in the two organic groups than in the conventional group. The 18:1 trans-10/18:1 trans-11 ratio was lower in the breastmilk collected from the women with high dairy fat intake than in the women with lower dairy fat intake. Similarly, 18:1 trans-10/18:1 trans-11 ratio was significantly lower in the groups consuming organic meat and dairy products than in the group, which consumed conventional diet.

### 3.3.3 COPPER AND ZINC

Mark *et al.* (2013) report findings of two double-blind, crossover trials, which investigated the intake and absorption of zinc and copper through diets based on organic growing system with animal manure, organic growing system with cover crops, and conventional growing system. Nine crops were cultivated to prepare diets for the study subjects. In 2008, 17 healthy non-smokers, mean age 24, average BMI was 22, and in 2009, 16 non-smokers, mean age 28, average BMI was 24 participated in the trials. Special diet menus were prepared. There were three dietary periods of 12 days in each trial. Intervention trial from day one to day seven was used as equilibration period. On day eight, stable enriched isotope labelled breakfast, lunch and dinner were served to the subjects. The last four days were used for analytical tracing of the isotope. The regular diets were compensated with 1mg ZnCl<sub>2</sub>/10 MJ from day one to day seven and from day nine to day 12. On day eight, ZnCl<sub>2</sub> was replaced with stable zinc isotope. The washout period between intervention periods was 14 days. Faecal samples were collected for 24 hours at baseline, two days before and after the isotope administration for the measurement of copper and zinc isotope. There was no difference in bioavailability of copper or zinc between organic or conventional diets.

### 3.3.4 PESTICIDES

Lu *et al.* (2006) reported that the consumption of organic diets decreased the urine concentration of organophosphorus pesticide metabolites in children. Twenty-three children, age 3-11 years, consumed conventional diet from day one to day three (phase 1), organic diet from day four to day eight (phase 2) and conventional diet, from day nine to day 15 (phase 3). Urine samples were collected every day, twice a day for 15 days. Among the five pesticide metabolites analysed, the urine concentration of pesticide malathion and chlorpyrifos metabolites, malathion dicarboxylic acid and 3,5,6-trichloro-2-pyridinol, were significantly decreased in the children during the phase 2 compared to phase 1 or phase 3. It was concluded that organic diets protected children from organophosphorus pesticide exposure.

Curl, Fenske and Elgethun (2003) estimated the level of organophosphorus pesticide exposure to preschool children from diets by measuring five pesticide metabolites in the urine samples. The age of the children ranged from 46 to 47 months, 56% were boys, all from Seattle, Washington, USA. Based on the label information on the food packs, 18 children consumed organic diets (≥75% organic foods) and 21 children consumed conventional diets, (≥75%

conventional foods). Age, sex, child activity and family economy were not significantly different between the two groups. Dimethylthiophosphate was the major metabolite detected in 87% of the urine samples. The children consuming organic foods had lower concentration of total dialkylphosphate metabolites, dimethylphosphate, dimethylthiophosphate, dimethyldithiophosphate, in the urine than in the children consuming conventional foods. Residential pesticide (pesticide residues in home) had no effect on the findings. It was suggested that the children consuming conventional foods might have been exposed to higher dimethyl organophosphorus pesticide exposure than the children consuming organic foods.

### 3.3.5 DISCUSSION AND SUMMARY OF THE MAIN FINDINGS IN BIOMARKER STUDIES

Studying biomarkers to evaluate possible effects on human health is a well-known but rather complex approach, and few well designed studies investigating effect of organic foods compared to conventional foods were found.

There are indications that increased oxidative stress is associated with chronic diseases. Data based on many studies suggest that increased bioavailability of antioxidants attenuate oxidative stress and may protect or delay onset of disease. Indications from plant analyses indicate somewhat higher concentrations of bioactive plant metabolites in organically grown products compared with conventionally grown, and it has therefore been hypothesised that the antioxidants capacity in blood would be increased in individuals who consume organic plant foods compared with individuals consuming conventional plant foods. In all but one study, organically grown foods did not result in a higher blood antioxidant capacity compared with conventionally grown foods (Di Renzo *et al.*, 2007). In one study it was found that antioxidant quercetin concentration in the organically produced foods was significantly higher than in the conventionally produced foods (Grinder-Pedersen *et al.*, 2003). However, the consumption of the organically produced foods resulted in a higher urinary excretion of the metabolites quercetin and kaemferol. One explanation for these findings might be that abundant intake of antioxidants in an experimental setting may only lead to an increased excretion in health subjects with sufficient intake, i.e. the cup is full. It is possible that higher antioxidant concentrations with use of organic food would be seen in energy and nutrient deficient subjects, as was indicated in the animal study by Finamore *et al.* (2004).

Zinc and copper are cofactors in several antioxidant enzymes and the result from the study of Mark *et al.*, (2013) showed no difference in intake or absorption of zinc or copper with use of an organic diet as compared with conventional diets.

The concentrations of trans fatty acid isomers (CLA and trans-vaccenic acid) were altered in breastmilk in women who consumed diets composed of >90% organic meat and dairy products (Rist *et al.*, 2007 and Mueller *et al.*, 2010). Increased concentration of these trans fatty acids is reported to have anti-inflammatory and growth supporting properties, and hence a positive effect on a child's health could be expected. It remains, however, to be established whether increased intake of these trans fatty acid implicate health benefits in new born children.

Two studies from USA showed a significant reduction of urinary pesticides excretion in children during dietary periods with mostly organic food as compared with conventional foods. Similar studies have not been conducted in Norway or in any of the Nordic countries, but in general exposure to pesticides in the Nordic countries were presumed to be lower than in USA and lower excretion of pesticide metabolites would be expected. However, Norwegian pregnant women participating in the Norwegian Mother and Child cohort Study had higher total urinary concentration of organophosphate pesticide metabolites compared to

pregnant women participating in the National Health and Nutritional Examination Survey in USA (Ye *et al.* 2009). The higher concentration of pesticides in Norway might be explained by that about 98% of fruits and 69% of berries sold in Norway are imported<sup>2</sup>.

To summarise, organic and conventionally produced foods resulted in similar antioxidant capacity in healthy subjects. There are some indications of increased concentration of anti-inflammatory and growth stimulating trans fatty acids in human milk from mothers using predominantly organically produced dairy and meat products. Children consuming an organic diet as compared to a conventional diet had lower urinary concentrations of pesticide metabolites.

### 3.4 OTHER STUDIES - CELL PROLIFERATION - *IN VITRO*

This section contains a study that couldn't be categorised as human studies, animal model studies or biomarker studies. The study investigates cell proliferation status, and the reported findings may be indirectly related to human health and organic foods.

In their study, Olsson *et al.* (2006) used extracts from five different strawberry-types grown either conventionally or in an organically in one Swedish research station. They used a measuring system based on suppression of proliferation of human colonic and breast cancer cell lines. The highest concentration of the organically-derived extracts caused a more pronounced antiproliferative effect (in the order of 10% better) than extracts obtained from conventionally grown strawberries.

### 3.5 METHODOLOGICAL ASPECTS - LIMITATIONS

During the preparation of this report several methodological challenges in studies investigating organic food and the effect on human health became evident. A frequent problem was noted with regard to the inadequate descriptions of the foods which were tested in the various studies, also in terms of production conditions. The chemical composition of plant products is determined by multiple factors, e.g. cultivar variety, soil conditions, precipitation, and amount of light. All these conditions have to be controlled for if the result of the agricultural system is to be evaluated properly.

Another major problem identified in the literature was the lack of a suitable design, both for experimental studies, but perhaps even more so for clinical trials. Clearly, the most robust design would be to embark on a placebo-controlled, double-blinded, randomised trial. Ideally, such a trial should be sufficiently powered to address clear and meaningful primary clinical endpoints, e.g. risk of various diseases or mortality. An adequately selected control-group is mandatory. We have not been able to identify any study in our literature search that satisfies these criteria. However, such trials are suited for medicines or food supplements, while interventions with whole diets cannot be run over months or years while waiting for eventual health outcomes to appear. Carefully conducted studies with other designs e.g. observational epidemiological studies (preferably prospective) are therefore realistic alternatives. In addition to clinically relevant endpoints, important knowledge might also be collected via sampling of various nutrient and non-nutrient biomarkers and/or other compounds (e.g. pesticides and other contaminants). Although meaningful endpoints should be fairly easy to

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<sup>2</sup>Totaloversikten; Frisk frukt, bær, grønnsaker og poteter 2003-2012. Oppdatert 19. mars 2013.

identify for a clinical trial, the choice of biomarkers is probably more difficult. With regard to experimental studies, e.g. concerning cells cultured *in vitro*, there is a need for standardisation of the experimental conditions (types of cells, dose and frequency of adding organically prepared solutions etc.).

Animal studies may add to the knowledge about the possible positive or negative effects of organic foods. The included animal studies were well-designed and investigated relevant endpoints and selection of biomarkers. However, the extrapolation from animal studies to humans remains a challenge. This might be particularly relevant to the field of possible benefits of organic food on human health. Rodents, the species most often used as experimental animals, display quite different characteristics in terms of e.g. life span, metabolic rate, and immune responses, compared with humans. Moreover, chronic disorders rooted in a sedentary lifestyle and/or poor diets are quite rare among laboratory animals in general.

To summarise, the present report has uncovered shortcomings in experimental design and reporting and consequently identified a marked need for more and improved studies in order to establish whether consumption of organic food is beneficial for human health. It is of importance to gain more knowledge both on eventual positive and negative health effects.

## 4 Reviews

The results and conclusions in the reviews described in this chapter are mainly based on the same studies as included in our chapter 3. The results and conclusions from these reviews are therefore not taken into consideration in our conclusions in chapter 7. The description of findings in the reviews is included as they give an overview of the scientific literature in this field and lines will be drawn to compare the findings in these reviews with our own results and conclusions. A short description of the aim of the reviews, number of included studies and main conclusions are given below. In section 4.1 additional specific human health outcomes and reported differences in concentrations of nutrients, other bioactive compounds and contaminants between organically and conventionally produced foods are outlined.

A total of 12 reviews addressing possible health outcomes related to consumption of organically produced food were found, of which two were based on statistical meta-analyses (Brandt *et al.* 2011, Smith-Spangler *et al.* 2012). The reviews differ in their aims, and in the types of studies included. Brandt *et al.* (2011) assessed only fruit, vegetables and herbs. Huber *et al.* (2011) and Velimirov *et al.* (2010) had a particular focus on animal studies. The number of studies directly addressing possible effects of organic food on human health is low, and as a consequence, conclusions in reviews (in particular the older ones) are mostly based on evaluations of differences in contents of nutrients or other substances of relevance for health. In one review, an estimation of the impact on human health, in terms of life time expectancy, of a higher content of secondary plant metabolites in organic food is calculated (Brandt *et al.* 2011).

Smith-Spangler *et al.* (2012) included 17 studies in humans and 223 studies of nutrient and contaminant levels in foods in their review. Their aim was to review evidence comparing the health effects of organic and conventional foods. Of the 17 studies in humans, three examined clinical outcomes, one reported *Campylobacter* infections, and the remaining studies examined health markers such as serum lipid or vitamin levels. The authors described the reviewed studies as generally heterogeneous and limited in number, and that publication bias may be present. They concluded that the reviewed literature lacks strong evidence that

organic foods are significantly more nutritious than conventional foods, but that consumption of organic foods may reduce exposure to pesticide residues and antibiotic-resistant bacteria (Smith-Spangler *et al.* 2012).

The review by Brandt *et al.* (2011) had two aims: 1) to perform a meta-analysis of the published comparisons of the content of vitamins and other bioactive metabolites in organically and conventionally produced fruits and vegetables, and 2) to estimate the potential increase in life expectancy that would be achieved by switching from conventional to organic fruits and vegetables without changing the amount consumed per day. A total of 102 papers were included, and a quantitative meta-analysis was performed. The reported result was that the content of bioactive metabolites in organic foods is on average 12% higher than in corresponding conventional samples, an overall difference spanning a large variation among sub-groups of secondary metabolites, from a 16% higher content for defense-related compounds to a non-significant 2% lower content for carotenoids, while vitamin C showed a 6% higher content. Based on the assumption that increasing the content of biologically active compounds in fruits and vegetables by 12% would be equivalent to increasing the intake of fruits and vegetables by the same 12%, it was estimated that the potential increase in life expectancy that would be achieved by switching from conventional to organic fruit and vegetables without changing the amount consumed per day, to 17 days for women and 25 days for men. The model developed by Brandt *et al.* (2011) to perform this estimation was based on a model by (Veerman *et al.* 2006) to estimate changes in lifestyle expectancy caused by changes in fruit and vegetable intake. To put these results in perspective, the authors referred to screening for breast cancer which has been calculated to provide an average increase in life expectancy of 35 days (Bonneux 2003).

The review by Dangour *et al.* (2010) (commissioned by the UK Food Standards Agency), included 12 studies and was aimed at assessing the strength of evidence that nutrition-related health benefits could be attributed to the consumption of foods produced under organic farming methods. It included eight human studies (six clinical trials, one cohort study, and one cross-sectional study) and four animal studies or human cell lines or serum. Dangour *et al.* concluded that the majority of the studies showed no evidence of differences in nutrition-related health outcomes that result from exposure to organic or conventionally produced foodstuffs, and that evidence is lacking for nutrition-related health effects that result from the consumption of organically produced foods. The authors reported that a quantitative meta-analysis was not considered justified due to the scarcity of studies and heterogeneity with regard to study designs, exposures tested and health outcomes investigated (Dangour *et al.* 2010).

The review by Guéuen and Pascal (2010) is an update of the report published in 2003 by the French Agency for Food Safety (AFSSA) a nutritional and safety evaluation of organically produced foods. Approximately 100 studies were published since the last review in 2003. The main part of the review concerned differences in the contents of nutrients and other bioactive compounds, such as secondary plant metabolites, nitrate and pesticide residues. The authors concluded that the small differences observed in chemical composition between organic and conventional foods have no practical significant effect on nutrition and health.

The aim of the review by Huber *et al.* (2011) was to give an overview of recent studies investigating the health value of organic foods and to present a framework for estimating the scientific impact of these studies, while some studies in humans were also reviewed. The authors concluded that if “health effects” are defined as effects on defined diseases in humans, evidence for such effects is presently lacking. Based on the literature, a hypothesis is proposed that organic food increases the capacity of living organisms to recover to

homeostasis – referring to the term ‘resilience’ – suggested to reflect a qualitatively good state of health, and to confirm this, effect studies on specific markers for health are called for.

The aim of the review by Velimirov *et al.* (2010) was to describe the state of the art of animal experiments used for comparing the impact on the animals' health as a model for the effects experienced by the human consumers. A total of 18 studies were reviewed, including plant-derived feeds only. These studies included feeding experiments with different fertiliser applications; feeding experiments with feed from organic versus conventional growing systems; feeding studies with the aim of defining biomarkers for health as proxy for human health, measuring differences between organic and conventional diets. The authors reported that the majority of the reviewed feeding experiments revealed effects of the organically produced feed on health parameters such as reproductive performance and immune responses. They concluded that the science-based results suggest positive influences from organic feeds, but there is still a need for confirmation in animals and, finally, in humans. The authors remarked that for this purpose animal feeding trials with feed from different production systems should be conducted, with the aims to define health indicators and to establish biomarkers as a basis for future dietary intervention studies in humans.

The review by Lima and Vianello (2011) aimed at reporting on the main characteristics and properties of plants cultivated organically and conventionally, and concluded that intake of organic foods appears to lead to some advantages, such as higher content of phenolic compounds and some vitamins, such as vitamin C, and a lower content of nitrates and pesticides.

Crinnion (2010) mainly reviewed studies comparing organically vs. conventionally grown foods with regard to contents of vitamins, minerals, phytochemicals and pesticide residues, as well as one study on allergic manifestations in children. The author concluded that although *in vitro* studies of organic fruits and vegetables consistently demonstrate that organic foods have greater antioxidant activity, are more potent suppressors of the mutagenic action of toxic compounds, and inhibit the proliferation of certain cancer cell lines, *in vivo* studies of antioxidant activity in humans have so far not demonstrated additional health benefits. It was further concluded that health benefits from consuming organic dairy products have been demonstrated in regard to allergic dermatitis.

Holmboe-Ottesen (2004) reported that organic foods compared to conventionally produced foods seem to have a higher content of antioxidants in plant products and a higher content of fat-soluble vitamins and n-3 fatty acids in animal products. Animal studies show higher fertility and less morbidity in animals fed organically. She further reported that no controlled studies in humans have assessed health effects, although a higher content of antioxidants have been found in the urine of subjects put on an organic diet. However, she concluded that a change to an organic diet may have a direct positive health effect on humans, as well as an indirect effect through a healthier environment.

Magkos *et al.* (2003), reported that there is little evidence that organic and conventional foods differ in respect to the concentrations of the various micronutrients (vitamins, minerals and trace elements), but that there seems to be a slight trend towards higher ascorbic acid content in organically grown leafy vegetables and potatoes. They also reported a trend towards lower protein concentration but of higher quality in some organic vegetables and cereal crops. With respect to the rest of the nutrients and the other food groups, the authors found that existing evidence is inadequate to allow for valid conclusions. Animal feeding experiments indicated that animal health and reproductive performance were slightly improved when they are organically fed, but a similar finding has not yet been identified in humans. They concluded

that despite any differences, it seems that a well-balanced diet can equally improve health regardless of its organic or conventional origin (Magkos, Arvaniti and Zampelas 2003a).

In their review on food safety aspects of organically vs. conventionally produced foods, Magkos *et al.* (2003), reported that some organic foods can be expected to contain fewer agrochemical residues and lower levels of nitrate than conventionally grown alternatives. On the other hand, environmental contaminants are equally present in foods of both origins. With regard to other food hazards, such as natural chemicals, microbial pathogens and mycotoxins, no clear conclusions could be drawn, although several interesting points were highlighted. They concluded that it is difficult to weigh the risks, but remarked that it should be made clear to consumers that "organic" does not equal "safe" (Magkos, Arvaniti and Zampelas 2003b).

In 2006, Magos *et al.* presented a critical discussion of issues that relate to the safety of organically and conventionally produced food. They called for caution in making firm conclusions based on the literature available at that point, and they also discussed the wider context of food quality which may be perceived as relevant for consumers when they consider organic vs. conventional food in the market (Magkos, Arvaniti and Zampelas 2006).

#### **4.1 HEALTH OUTCOMES AND COMPARISON OF NUTRIENTS, OTHER BIOACTIVE COMPOUNDS AND CONTAMINANTS DISCUSSED IN THE REVIEW**

In the study by Kummerling *et al.* (2008) addressed in Smith-Spangler *et al.*, 2012; Huber *et al.*, 2011; Crinnion, 2010 and Dangour *et al.*, 2010, it was reported that consumption of dairy products of which more than 90% were organically produced lowered the risk of eczema in children. The study by Kummerling *et al.* is included in our section 3.1.1.

One study of differences in sperm quality related to consumption of organic vs. conventional food was reviewed by Smith-Spangler *et al.* (2012), who concluded that no clinically meaningful differences were found. This study is among the studies included in our section 3.1.2.

In studies investigating nutrient concentrations in organic vs. conventionally produced foods, higher levels of vitamin C and phosphorous and a tendency towards lower levels of carotenoids were reported. It was further reported a trend towards lower protein concentration but of higher quality in some organic vegetable and cereal crops. However, most reviews concluded that these differences are of no clinical significance. The authors concluded that higher contents of some types of secondary plant metabolites have also been demonstrated in organically grown produce compared with conventionally grown. However, it remains to be shown whether the level of differences between organically and conventionally produced food have health-related implications (Brandt *et al.*, 2011; Crinnion, 2010; Gueguen and Pascal, 2010, Holmboe-Ottesen, 2004; Huber *et al.*, 2011; Lima and Vianello, 2011; Magkos *et al.*, 2003a and Smith-Spangler *et al.*, 2012). These conclusions are based partly on the same studies as included in our section 3.3.1, while some of the reviews have investigated a large number of studies on differences in levels of nutrients in organic vs conventional foods not included in our report.

Higher concentrations of n-3 fatty acids and/or conjugated linoleic acid in milk and meat from organically raised animals were reported in Crinnion, 2010; Gueguen and Pascal, 2010; Holmboe-Ottesen, 2004; Huber *et al.*, 2011 and Smith-Spangler *et al.*, 2012. The two studies of Rist *et al.* (2007) and Mueller *et al.* (2010) where higher concentrations of conjugated linoleic acid and trans-vaccenic acids were found in the breastmilk of mothers who reported consumption of a strictly organic diet were addressed in Crinnion, 2010; Drangour *et al.*,

2010; Huber *et al.*, 2011 and Smith-Spangler *et al.*, 2012. These findings are partly based on the same studies as included in our section 3.3.2.

Lower levels of dietary exposure of pesticide residues, measured as pesticide metabolites with use of organic diets compared to conventional diets were commented on and all reviewers concluded that consumption of organic food provides protection against exposure to organophosphorous pesticides commonly used in agricultural practices (Crinnion, 2010; Gueguen and Pascal, 2010; Holmboe-Ottesen, 2004; Huber *et al.*, 2011; Lima and Vianello, 2011; Magkos *et al.*, 2003b and Smith-Spangler *et al.*, 2012).

With regard to mycotoxins, equal or lower amounts were reported in organic crops compared with conventional crops (Gueguen and Pascal, 2010; Huber *et al.*, 2011; Lima and Vianello, 2011; Magkos *et al.*, 2003b and Smith-Spangler *et al.*, 2012). Lower levels and lower risk for contamination with deoxynivalenol (DON) was found in organic grains than conventional alternatives (Smith-Spangler *et al.*, 2012).

It is reported that estimates of differences in levels of heavy metal contamination in food are highly heterogeneous, and it is generally concluded that these substances may be equally present in food from both farming systems (Magkos *et al.* 2003b and Smith-Spangler *et al.*, 2012). No significant differences in cadmium or lead content were identifiable (Smith-Spangler *et al.*, 2012).

Several studies report lower levels of nitrate in organic vs. conventional foods (Holmboe-Ottesen, 2004; Huber *et al.*, 2011; Lima and Vianello, 2011; Magkos *et al.*, 2003b and Magkos *et al.*, 2006).

With regard to bacterial infections it is generally reported similar risks independent of farming system (Magkos *et al.*, 2003b and Magkos *et al.*, 2006). An exploratory case-control study showed no significant differences in symptomatic *Camphylobacter* infections between organic and conventional animal products (Smith-Spangler *et al.*, 2012). It was reported that *Escherichia coli* contamination risk did not differ between organic and conventional produce. Bacterial contamination of retail chicken and pork was common but unrelated to farming method. However, the risk of isolating bacteria resistant to three or more antibiotics was higher in conventional than organic chicken and pork (Smith-Spangler *et al.*, 2012).

To summarise, the overall conclusion of these reviews is that no clear effects on human health have been presented as a result of consuming an organic diet compared to a conventional diet. However, reduced frequency of atopic disease among children growing up with consumption of organic milk was reported.

In one review, a 12% increase in intake of biologically active compounds with consumption of organically grown fruits and vegetables was suggested. This was evaluated to be equivalent to a 12% increase in intake of fruits and vegetables and was modelled to represent a small increase in life expectancy. Some other reviews also commented on higher concentrations of biologically active compounds with an organic diet compared to conventional and related this to human health impact, while others concluded that the small differences in intake would not have health consequences for humans.

A reduced pesticide exposure with organic diet was commented on in several reviews. However, since no human studies had been conducted to verify a positive effect of the lower pesticide exposure no conclusions could be made on health impact in humans.

Well-powered human studies for future research is called for and partly outlined.

## 5 Reports prepared by governmental bodies

Other European governmental bodies have prepared reviews and conducted assessments of the scientific knowledge on organic foods, nutrition and health. The mode of working and main conclusions from two governmental reports will be presented here.

### 5.1 FOOD STANDARDS AGENCY, UNITED KINGDOM

The British Food Standards Agency commissioned the London School of Hygiene & Tropical Medicine to write the report “Comparison of putative health effects of organically and conventionally produced foodstuffs: a systematic review” in 2009 (FSA 2009).

Background: The large and increasing demand for organic foods in the UK and elsewhere.

Literature search: Systematic literature review covering peer-reviewed journals between 1958 and 2008. Only three of the 11 included studies met the pre-defined satisfactory quality criteria.

Aim: To systematically review the evidence of differences in putative health effects of organically produced compared with conventionally produced foodstuffs. Focus on the nutritional content of foods. The contaminant content or the environmental impact of the agricultural practises was not addressed.

Type of studies: Six human studies including four clinical trials; five *in vitro* or *ex vivo* studies on human cell lines or serum.

Exclusion criteria: not peer-reviewed, no English abstract, direct comparisons was not possible, animal health was investigated from a veterinary perspective and not as a model for human health, were primarily concerned with content of other bioactive compounds.

Comparator: Conventional farming methods

Grading of evidence: No, not possible because of the high diversity of the included studies.

Main conclusions: Because of the limited and highly variable data available, and concerns over the reliability of some reported findings, there is currently no evidence of a health benefit from consuming organic compared to conventionally produced foodstuffs. It should be noted that this conclusion relates to the evidence base currently available on the nutrient content of foodstuffs.

### 5.2 AGENCY FOR FOOD, ENVIRONMENTAL AND OCCUPATIONAL HEALTH AND SAFETY, FRANCE

The French Food Safety Agency (Agence Française de Sécurité Sanitaire des Aliments) published the report “Nutrition and health assessment of organic food” in 2003 (AFSSA 2003).

Background: An increase in production and consumption of organic foods in France. No overall evaluation had been conducted on the specific practices of organic agriculture in terms of nutrition and health. The working group consisted of both scientists and stakeholders.

Aim: Gather scientific data and perform a benefit and risk assessment.

Scientific literature search: Emphasis on articles published since 1980 and published internationally. Clear inclusion and exclusion criteria. When data were missing, a deductive approach was used.

Type of studies: Comparative studies, i.e. organic vs conventionally produced foods.

Exclusion criteria: Unknown history of the farmland, incorrect practices in terms of organic standards, non-validated data.

Comparator: Conventional farming methods.

Grading of evidence: No.

Main conclusions:

- In terms of nutrition, no conclusion could be drawn as to differences in nutrient content of organic foods and foods produced in a conventional manner. Slight tendency to higher dry matter content in organically produced vegetables, but not fruits. Slightly lower protein content in organically produced cereals. Better amino acid balance. Notable differences in fatty acid profiles in animal products. Slightly higher vitamin C in organically produced potatoes, no differences in  $\beta$ -carotene in vegetables. The only significant differences found were the higher content of polyphenols in organic fruit and vegetables.
- The assessment notes that some technological processes used in organic farming practices are likely to have consequences for nutritional quality of the foods. For example, the milling processes enable better preservation of the germ and bran in the flour, resulting in breads richer in minerals, fibre and vitamins.
- In terms of health and food safety, the majority of studies show no residues of pesticides in organic foods. As to mycotoxins, no major differences were reported. Lower nitrate levels in organically produced foods. Dioxins and other environmental pollutant exposure happen as a result of contamination and are not specific to any particular agricultural system.

## 6 Comments on differences between conventional and organic foods - nutrients, other bioactive compounds and contaminants

This chapter will briefly discuss eventual differences in the content of substances (nutrients and other bioactive compounds, pesticides, other contaminants and mycotoxins) in food products from the two production systems. Several of the substances have partly been covered by sections in the VKM opinions Part I: Plant health and plant production and Part V: Pesticide residues.

### Nutrients and other bioactive compounds (secondary metabolites) in plants

#### *Nutrients*

The VKM opinion Part I: Plant health and plant production has given the following conclusion regarding comparison of nutrient and other bioactive compound content in organically or conventionally produced food: "The evaluation shows that protein content is commonly lower in organically produced wheat than in conventional wheat. Dry matter and starch contents are higher in organic than in conventionally produced potato. Higher nutrient levels in the soil support rapid growth of the potato tuber at the expense of dry matter and starch content in conventional farming. In most studies the nitrate content is higher in conventional than in organic potato due to higher soil nitrogen availability in conventional than in organic farming.

Most studies report higher levels of dry matter, ascorbic acid and antioxidant activity in organic than in conventional apples. Organic grown berries may have higher sensory quality and content of secondary plant metabolites with antioxidant activity and some minerals than conventional berries.

For vegetables the results were variable and less clear. In some studies vegetables grown in organic production systems have higher content of some nutrients and secondary plant metabolites with antioxidant activity, while in other studies there is no effect of the growing system."

Based on the conclusions from the Part I: Plant health and plant production, results from comparative studies do not allow for firm conclusions as to major differences in nutrient content between conventionally and organically grown plants. Fertilisers with high nitrogen content drives the water content of plants up somewhat and leads to a certain dilution of nutrient concentrations, which explains the slightly lower content of some nutrients in conventionally grown products. Oppositely, a higher nitrogen and nitrate content have been found in many studies of conventional compared with organically grown cereals and vegetables explained by the high nitrogen input from artificial fertilisers.

#### *Other bioactive compounds (secondary plant metabolites)*

Higher contents of secondary plant metabolites have been reported in organically compared to conventionally grown foods in the Part I: Plant health and plant production, and in several single studies and reviews included in this report. This has been explained in the context of restrictions in the type and intensity of fertilisation in organic agriculture (Brandt and Molgaard 2001, Brandt *et al.* 2013).

In one review, a 12% increase in intake of biologically active compounds with consumption of organically grown fruits and vegetables was suggested. This was evaluated to be equivalent to a 12% increase in intake of fruits and vegetables and was modelled to represent a small increase in life expectancy.

Some other reviews also commented on higher concentrations of biologically active compounds with an organic diet compared to conventional and related this to human health impact, while others concluded that the small differences in intake would not have health consequences for humans. The results vary with regard to different types of secondary plant metabolites, and it remains to be shown whether the level of differences between organically and conventionally produced food have health related implications.

## **Pesticides**

Both animal and human studies have shown that pesticides may have a negative impact on reproductive health (Fernandez *et al.* 2007, WHO/UNEP 2013). Synthetic pesticides are not permitted in organic farming, and lower concentrations of pesticides have been documented in organically grown compared with conventional grown vegetables, fruits and berries in the VKM opinion Part V: Pesticide residues and in several of the included single studies and reviews included in this report. Data from the 2010 EFSA Report showed that 4.2% of the conventional and 1.0% of the organic plant products exceeded the MRL values (maximum residue level), and 43.3% of the conventional and 10.8% of the organic plant products had measurable residues below the MRL (EFSA 2013). Of 624 organic samples analysed in Norway over the last six years only one sample had residues exceeding MRL.

The main conclusion from the VKM opinion Part V: Pesticide residues is that the general levels of pesticide residues in both conventional and organic food are low, and well below what is likely to result in adverse health effects. Furthermore they conclude: "Although the data in part is insufficient to perform quantitative comparisons of residue levels between conventional and organic products, it is concluded that organic food contains far lower amounts of pesticides than conventional food. No generally accepted methodology is at present established for cumulative risk assessment of combined exposure to pesticide residues. Available data suggest however that combined exposure is not likely to result in increased human health risk."

Although organic food not seems to be completely free from pesticide residues, due to contamination from conventional agriculture or fraud, organic foods have been convincingly demonstrated to expose consumers to fewer and lower levels of pesticide residues. As discussed in section 3.3.5 in this report, the importance of lower pesticides exposure with organic compared to conventionally produced food on human health has to be investigated further.

### **Other contaminants**

The VKM opinion Part I: Plant health and plant production has given the following conclusion regarding comparison of contaminants content in organically or conventionally produced food: "The uptake in plants of most organic chemical contaminants from soil is very low or negligible. Differences in organic contaminants in soil are probably mostly related to sources that are not influenced by organic and conventional practices. Due to high human consumption, cereals and vegetables are important sources for dietary intake of heavy metals. The data do not provide basis for a conclusion on differences in levels of metals between organically and conventionally grown food plants."

This is in line with the reviews included in this opinion, which also says that environmental contaminants are equally present in foods of both origins, and that differences in levels of heavy metal contamination contaminants in food are highly heterogeneous. It is generally concluded that these substances may be equally present in food from both farming systems. Consequently, no conclusions can be drawn regarding health effects related to contaminant differences.

### **Mycotoxins**

The VKM opinion Part I: Plant health and plant production has given the following conclusion regarding comparison of contaminants content in organically or conventionally produced food "Contamination of cereals with *Fusarium* mycotoxins is widespread. Results from comparison of mycotoxin contamination in organic and conventional cereals are variable. Most studies found no difference in DON content and the majority of the remaining studies reported on lower levels in organic than in conventional cereals. Most studies showed that organically produced cereals contained lower levels of T-2 and HT-2 toxin than conventionally grown cereals. Organic cereal farmers practice wider crop rotation, more ploughing, and they apply less fertiliser which gives lower plant density then on conventional farms. DON producing fungi are partly controlled by fungicides in conventional farming, while there are no approved fungicides for control of T-2 and HT-2 producing fungi. Some studies showed higher mycotoxin contamination in organic than in conventional apple products, while other studies reported similar contamination. The difference may be due to

more efficient disease control in conventional orchards, which reduces the mycotoxin producing fungi in apple fruits."

The similar or lower levels of trichotecene mycotoxins (DON, T-2 and HT-2) in organic compared with conventional produced cereals may be of importance for infants and children who have a high consumption of cereals (VKM 2013).

### **Differences in milk and dairy products**

In two articles, one from Norway in 2003 and in a more recent study from UK in 2012, organic milk was found to have lower concentrations of iodine than conventionally produced milk.

In Dahl *et al.* (2003) results of iodine concentration in Norwegian-produced milk and in a selection of dairy products was reported. Low-fat conventional and organic milk were sampled from nineteen and seven different locations in Norway, respectively, during the summer and winter season of 2000. The median iodine concentration of organic summer milk (60 µg/l) was significantly lower than the iodine concentration of organic winter milk (127 µg/l) and lower than the corresponding conventionally produced milk (88 µg/l and 232 µg/l respectively). The variance (35 - 365 µg/l in winter milk) was greater in the organic produced milk compared to the conventionally produced milk (103 - 272 µg/l), which might indicate a more inconsequent use of iodine supplementation of organic fodder or that cows in organic farms have a longer out-of-doors season. There were no significant differences in iodine concentration with respect to geographical sampling location. With a recommended iodine intake of 150 µg/day for adults, a daily intake of 0.4 liters milk meets the requirement with 16% during the summer and 34% during the winter season with use of organic milk as compared to 25% and 60% with conventional milk.

Bath, Button and Rayman (2012) aimed to compare the iodine concentration of retail organic and conventional milk and to evaluate regional influences in iodine levels in the UK. Samples of organic milk (n=92) and conventional milk (n=80), purchased from retail outlets in sixteen areas of the UK (southern England, Wales and Northern Ireland), were analysed for iodine. It was shown that the iodine content was 42.1% lower in organic milk than conventional milk (median iodine concentration 144.5 v. 249.5 µg/l; p<0.001). There was no difference in the iodine concentration with regard to area of purchase, but a difference was seen in iodine concentration of organic milk by region of origin (p<0.001).

The Norwegian study points at the importance of milk and milk products as sources of iodine. Based on information from TINE (personal information from Kristi Brønner 11.04.2014), the iodine concentration in organically produced milk has increased to 200 µg/l, and the concentration is now similar to the iodine concentration found in conventionally produced milk.

### **Differences in eggs**

Elevated concentrations of organic environmental pollutants as dioxins and PCBs have been observed in eggs from free range hens or eggs from foraging hens, relative to conventionally produced eggs (Schuler, Schmid and Schlatter 1997). The exposure of such compounds is thought to be increased in hens allowed access to the outdoors because they have increased access to additional sources of these compounds as soil, worms, insects and non-commercial feeds, relative to those held in cages (De Vries, Kwakkel and Kijlstra 2006). However, the main sources of PCBs and dioxins are oily and semi-oily fish and seafood, and increased

concentrations of PCB and dioxin in eggs are of no major importance to the total exposure of these contaminants (Kvalem *et al.* 2009).

To summarise, there are some differences in concentrations of nutrients and other bioactive compounds in organically compared to conventionally produced foods. However, the relevance for human health remains to be explored.

Pesticide exposure is lower with use of organic food compared to conventional food. The importance of lower pesticides exposure with organic compared to conventionally produced food on human health in a Norwegian setting has to be investigated further.

A possible lower concentration of mycotoxins with organic food production would be positive for human health and especially for child health.

No negative or positive effect on human health can be drawn based on differences in contaminant concentrations based on cultivation system.

## 7 Conclusions

Studies to investigate the health impact of eating organically produced foods compared to conventionally produced foods are so far scarce and have several limitations as discussed in section 3.5. In the literature search VKM found that the clinical health outcomes studied have been investigated in too few studies to allow for firm conclusions:

- Immune-associated outcomes, three studies – indications of a positive effect of organic food
- Semen quality, four studies – no conclusion can be drawn
- Hypospadias, only one study – no conclusion can be drawn
- Risk factors for cardiovascular diseases, only one study – no conclusion can be drawn

None of the included studies on human health reported negative health effects from organic food consumption compared with conventionally produced food. However, the studies were not specifically designed to investigate safety.

Animal models with feeding trials of food from different production systems have been investigated. Results from these studies might indicate a positive effect of organic feed on some outcomes; however, the extrapolation from animal studies to humans remains a challenge:

- Immune-associated outcomes, eight studies – no consistent findings
- Body composition, nine studies – no consistent findings
- General health, four studies – increased longevity and fertility in banana-flies on organic diet (one study), decreased daytime activity in rats on organic diet (one study), two studies showed no differences – different parameters investigated, and no conclusion can be drawn

For the studies on biomarkers, there was a lack of a clear operational definition of health. The inability to distinguish between different levels of health using valid biomarkers was among

the most striking short-comings. It was difficult to relate any biomarker to good health or protection against disease:

- Antioxidant capacity, eight studies – probably no differences
- Urinary pesticide concentrations, two studies in children – significantly lower concentration of urine pesticides after consuming organic diets compared to conventional diets
- Fatty acid concentration in breastmilk, two studies – higher concentration of trans fatty acids with organic diet compared to conventional

Based on national and relevant international research it has not been possible to draw firm conclusions about possible positive health effects of organically compared to conventionally produced foods. Indications of health benefits from organic food on risk of atopic diseases in children were found. Animal model studies indicated positive impact on general health of organic diets. However, no consistent findings were found which enable a conclusion. An organic diet was found to give higher concentrations of trans fatty acids (different from industrially produced trans fatty acids, i.e. vaccinic- and conjugated linoleic acid) in breastmilk of lactating women, and lower urinary concentrations of pesticide metabolites in children. The relevance of these findings for human health in Norway is uncertain. There are some differences in concentrations of nutrients and other bioactive compounds in organic in comparison with conventional foods. However, differences are small and the relevance for health in humans on a well-balanced diet is uncertain. The impact of organic diets on human health calls for more well designed and well powered studies. None of the studies on human health reported negative health effects from organic food consumption compared with conventional foods.

These findings are in line with several previous reviews which also conclude that no clear positive effects on human health have been presented as a result of consuming an organic diet in comparison with a conventional diet.

## 8 Data gaps

The work with this literature review has revealed a scarcity of relevant studies, and accordingly many data gaps. Research within the field shows lack of consistent findings and inconclusive results. It is of importance to gain more knowledge both on eventual positive and negative health effects.

### *Studies on human health*

An optimal study approach would be to conduct randomised controlled studies where two groups of humans ate the same diet, but the foods were grown according to organic farming in one group and conventional farming in the other group. A limitation would be that such studies can only be done for a shorter time period, e.g. some weeks or a month. Therefore, prospective observation studies can be used to study long-term associations between organic food consumption and health. Questions about organic food use should therefore be incorporated into food questionnaires in large cohort studies. Birth cohort studies and other cohorts with numerous participants like e.g. the EPIC or HUNT studies, would be valuable studies for collection of health impact of organic food consumption. A number of health

parameters could be studied, such as growth (in studies with children), immunological and neurological development, and risk of diabetes, coronary heart disease and cancer.

Randomised controlled studies will be important to achieve knowledge about important biomarker for human health.

Regarding mycotoxins, more work should be conducted on pregnant women and children since the foetus and small children are especially vulnerable to neurotoxicants because of their immature nervous systems, their rapid rate of brain growth and their low levels of the enzymes involved in the metabolism and detoxification of pesticides.

#### *Animal studies*

Animal experiments can be used to study doses and mechanisms of action. Some animal studies included in the present report indicate an effect of organic feed on immunity, longevity and fertility. This should be followed up with studies across several generations.

#### *Novel types of studies*

Organic farming will have an impact on soil microflora and accordingly, plant microflora. The diets impact on gut health is a rapidly expanding field of research. Further research in this field with focus on organic vs conventional food would open up new insight of the gut health impact on human health.

# Appendix 1

## Search 1

### Medline

1. organic\*.ti.
2. biodynamic\*.ti.
3. 1 or 2
4. food, organic/
5. organic food\*.ti,ab,sh.
6. organic diet\*.ti,ab,sh.
7. health food\*.ti,ab,sh.
8. 4 or 5 or 6 or 7
9. 3 and 8
10. limit 9 to (danish or english or french or german or norwegian or swedish)
11. letter.pt.
12. editorial.pt.
13. 11 or 12
14. 10 not 13
15. limit 14 to yr="1991 -Current"

### Embase

1. organic\*.ti.
2. biodynamic\*.ti.
3. 1 or 2
4. organic food/
5. organic food\*.ti,ab.
6. organic diet\*.ti,ab.
7. health food/
8. health food\*.ti,ab.
9. or/4-8
10. 3 and 9
11. limit 10 to (danish or english or french or german or norwegian or swedish)
12. conference abstract.pt.
13. editorial.pt.
14. letter.pt.
15. note.pt.
16. 12 or 13 or 14 or 15
17. 11 not 16
18. limit 17 to yr="1991 -Current"

## Search 2

### Medline

1. anthroposophy/
2. anthroposoph\*.ti,ab.
3. parsifal\*.ti,ab.
4. waldorf school\*.ti,ab.
5. waldorf education\*.ti,ab.
6. steiner school\*.ti,ab.
7. steiner education\*.ti,ab.
8. rudolf steiner\*.ti,ab.
9. or/1-8
10. Respiratory Tract Diseases/
11. Respiratory Tract Disease\*.ti,ab.
12. Arthritis/
13. arthritis.ti,ab.
14. Irritable Bowel Syndrome/
15. Irritable Bowel Syndrome\*.ti,ab.
16. Celiac Disease/
17. Celiac Disease\*.ti,ab.
18. Asthma/
19. Asthma\*.ti,ab.
20. Eczema/
21. Eczema\*.ti,ab,sh.
22. hypersensitivity/
23. hypersensiti\*.ti,ab.
24. sensiti?ation.ti,ab.
25. Pelvic Inflammatory Disease/
26. Inflammation/
27. Inflammatory Disease\*.ti,ab.
28. Avitaminosis/
29. Avitaminosis.ti,ab.
30. vitamin deficien\*.ti,ab.
31. Nutritional Status/
32. Nutritional Status.ti,ab.
33. Deficiency Diseases/
34. Deficiency Disease\*.ti,ab.
35. micronutrient deficien\*.ti,ab.
36. micro-nutrient deficien\*.ti,ab.
37. Reproductive Health/

38. Reproductive Health\*.ti,ab.
39. Reproduction/
40. reproduction\*.ti,ab,sh.
41. reproduction\*.ti,ab.
42. Fertility/
43. fertility.ti,ab.
44. Lactation/
45. lactation\*.ti,ab.
46. Breast Feeding/
47. breast feeding.ti,ab.
48. breastfeeding.ti,ab.
49. Semen/
50. semen.ti,ab.
51. Eye Diseases/
52. Eye Disease\*.ti,ab.
53. Obesity/
54. obesity.ti,ab.
55. Overweight/
56. overweight\*.ti,ab.
57. Weight Gain/
58. Weight Gain\*.ti,ab.
59. Weight Loss/
60. Weight Loss\*.ti,ab.
61. Adiposity/
62. adiposity.ti,ab.
63. weight change\*.ti,ab.
64. Body Weight/
65. body weight\*.ti,ab.
66. Body Mass Index/
67. body mass.ti,ab.
68. Eating Disorders/
69. Eating Disorder\*.ti,ab.
70. Anorexia Nervosa/ or Anorexia/
71. anorexia.ti,ab.
72. Bulimia Nervosa/ or Bulimia/
73. bulimia\*.ti,ab.
74. Binge-Eating Disorder/
75. binge-eating.ti,ab.

76. over eat\*.ti,ab.
77. Anthropometry/
78. anthropometr\*.ti,ab.
79. Body Composition/
80. Body Composit\*.ti,ab.
81. Body Constitution/
82. Body Constitut\*.ti,ab.
83. Waist Circumference/
84. Waist Circumference.ti,ab.
85. hip circumference.ti,ab.
86. Waist-Hip Ratio/
87. Waist-Hip Ratio.ti,ab.
88. Diabetes Mellitus, Type 2/
89. diabetes mellitus type 2.ti,ab.
90. type 2 diabetes.ti,ab.
91. diabetes type 2.ti,ab.
92. Insulin Resistance/
93. Insulin Resistan\*.ti,ab.
94. Hyperglycemia/
95. Hyperglycem\*.ti,ab.
96. Cardiovascular Diseases/
97. Cardiovascular Disease\*.ti,ab.
98. Coronary Disease/
99. Coronary Disease\*.ti,ab.
100. Osteoporosis/
101. Osteoporosis\*.ti,ab.
102. Bone Density/
103. bone density.ti,ab.
104. Neoplasms/
105. neoplasm\*.ti,ab.
106. cancer\*.ti,ab.
107. Tooth Diseases/
108. Tooth Disease\*.ti,ab.
109. Chronic Disease/
110. Chronic Disease\*.ti,ab.
111. Nervous System/
112. Nervous System\*.ti,ab.
113. neurodevelopment\*.ti,ab.

114. Mental Processes/
115. mental process\*.ti,ab.
116. Child Development/
117. Child Development.ti,ab.
118. Attention Deficit Disorder with Hyperactivity/
119. Attention Deficit Disorder\*.ti,ab.
120. adhd.ti,ab.
121. Autistic Disorder/
122. autism.ti,ab.
123. health outcome.ti,ab.
124. health measur\*.ti,ab.
125. birth outcome\*.ti,ab.
126. or/10-125
127. 9 and 126
128. limit 127 to yr="1991-Current"
129. limit 128 to (danish or english or french or german or norwegian or swedish)

## EMBASE

1. anthroposoph\*.ti,ab.
2. parsifal\*.ti,ab.
3. waldorf school\*.ti,ab.
4. waldorf education\*.ti,ab.
5. steiner school\*.ti,ab.
6. steiner education\*.ti,ab.
7. rudolf steiner\*.ti,ab.
8. or/1-7
9. respiratory tract disease/
10. respiratory tract disease\*.ti,ab.
11. arthritis/
12. arthritis.ti,ab.
13. irritable colon/
14. irritable colon\*.ti,ab.
15. irritable bowel\*.ti,ab.
16. celiac disease/
17. celiac disease\*.ti,ab.
18. asthma/ or allergic asthma/
19. asthma\*.ti,ab.
20. eczema/

21. eczema\*.ti,ab.
22. hypersensitivity/
23. hypersensiti\*.ti,ab.
24. sensitization/
25. sensiti?ation\*.ti,ab.
26. pelvic inflammatory disease/
27. inflammatory disease\*.ti,ab.
28. inflammation/
29. inflammation\*.ti,ab.
30. retinol/
31. vitamin a.ti,ab.
32. retinol\*.ti,ab.
33. vitamin deficiency/
34. vitamin deficien\*.ti,ab.
35. nutritional status/
36. nutritional status.ti,ab.
37. nutritional deficiency/
38. nutritional deficien\*.ti,ab.
39. deficiency disease\*.ti,ab.
40. micronutrient deficien\*.ti,ab.
41. micro-nutrient deficien\*.ti,ab.
42. reproductive health/
43. reproductive health\*.ti,ab.
44. reproduction/
45. reproduction\*.ti,ab.
46. fertility/
47. fertility\*.ti,ab.
48. lactation/
49. lactation\*.ti,ab.
50. breast feeding/
51. breast feeding.ti,ab.
52. breastfeeding.ti,ab.
53. sperm/
54. sperm\*.ti,ab.
55. semen.ti,ab.
56. eye disease/
57. eye disease\*.ti,ab.
58. obesity/

59. obesity.ti,ab.
60. overweight\*.ti,ab.
61. weight gain/
62. weight gain\*.ti,ab.
63. weight reduction/
64. weight reduction\*.ti,ab.
65. weight loss\*.ti,ab.
66. adiposity\*.ti,ab.
67. weight change\*.ti,ab.
68. body weight/
69. body weight\*.ti,ab.
70. body mass/
71. body mass\*.ti,ab.
72. eating disorder/
73. eating disorder\*.ti,ab.
74. anorexia nervosa/ or anorexia/
75. anorexia.ti,ab.
76. bulimia/
77. bulimia\*.ti,ab.
78. binge eating disorder/
79. binge eating\*.ti,ab.
80. over eat\*.ti,ab.
81. anthropometry/
82. anthropometr\*.ti,ab.
83. body composition/
84. body composit\*.ti,ab.
85. body constitution/
86. body constitut\*.ti,ab.
87. waist circumference/
88. waist circumference.ti,ab.
89. hip circumference/
90. hip circumference.ti,ab.
91. waist hip ratio/
92. waist hip ratio.ti,ab.
93. diabetes mellitus/
94. diabetes mellitus\*.ti,ab.
95. non insulin dependent diabetes mellitus/
96. diabetes type 2.ti,ab.

97. type 2 diabet\*.ti,ab.
98. insulin resistance/
99. insulin resistanc\*.ti,ab.
100. hyperglycemia/
101. hyperglycem\*.ti,ab.
102. cardiovascular disease/
103. cardiovascular disease\*.ti,ab.
104. coronary artery disease/
105. coronary artery disease\*.ti,ab.
106. coronary disease\*.ti,ab.
107. osteoporosis/
108. osteoporosis\*.ti,ab.
109. bone density/
110. bone density.ti,ab.
111. neoplasm/
112. neoplasm\*.ti,ab.
113. cancer\*.ti,ab.
114. tooth disease/
115. tooth disease\*.ti,ab.
116. chronic disease/
117. chronic disease\*.ti,ab.
118. nervous system/
119. nervous system\*.ti,ab.
120. neurodevelopment\*.ti,ab.
121. mental function/
122. mental function\*.ti,ab.
123. mental process\*.ti,ab.
124. child development/
125. child development\*.ti,ab.
126. attention deficit disorder/
127. attention deficit disorder\*.ti,ab.
128. adhd\*.ti,ab.
129. autism/
130. autism\*.ti,ab.
131. autistic disorder\*.ti,ab.
132. health outcome\*.ti,ab.
133. health measur\*.ti,ab.
134. pregnancy outcome/

135. pregnancy outcome\*.ti,ab.
136. birth outcome\*.ti,ab.
137. or/9-136
138. 8 and 137
139. limit 138 to yr="1991 -Current"
140. limit 139 to (danish or english or french or german or norwegian or swedish)
141. conference abstract.pt.
142. 140 not 141

## Appendix 2

### Human Studies

<b>Reference</b>	Alm, J. S., Swartz, J., Lilja, G., Scheynius, A., & Pershagen, G. (1999). Atopy in children of families with an anthroposophic lifestyle. <i>Lancet</i> , 353(9163), 1485-1488
<b>Study design and type</b>	Cross-sectional study in children with epidemiological and clinical parts.
<b>Objective</b>	To compare the prevalence of atopy in children from antroposophic families attending Steiner schools with children attending state schools.
<b>Number of participants, country and age</b>	295 children from Steiner schools, 380 children from state schools, age 5-13 years.
<b>Baseline characteristics of study subjects/cells</b>	Significant differences in the subjects from each group (Steiner schools vs state schools): Breastfeeding:> 4 months 85/65%, Antibiotics ever 52/90%, Any vaccination 91/100%, MMR 18/93, Measles history 61/1%, fermented vegetables 63/5%, organic/biodynamic food 76/6%.
<b>Exposure, substances, food (type and amount)</b>	Fermented vegetables (affecting intestinal microflora), other dietary factors not mentioned.
<b>Exposure; nature of the food (type and amount)</b>	An overall biodynamic/organic diet in children in Steiner schools (76%) vs 6% in state school children.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Atopy/ no atopy defined by history and/or clinical examination, skin prick test and blood test detecting sensitisation.
<b>Follow-up period, drop-outs</b>	One family in Steiner schools refused the clinical part of the study, 17 in state schools refused the clinical part of the study. These families are not included in the results. The clinical examinations took place between April and October 1997 by two physicians.
<b>Statistical analysis</b>	Odds ratio and 95% CI of atopy estimated by regression analyses. 95% CIs and p values (Wald test) based on an estimator of variance that relaxed the assumption of independence between observations within families.
<b>Results</b>	13% of the Steiner school children had a history of atopy or symptoms consistent with atopy, 25% of the children from state schools. Of all the children eating biodynamic/organic food 174 had no atopy while 54 had atopy.
<b>Conclusion</b>	There are many factors in the antroposophic lifestyle that may be preventive of atopy and organic vs conventionally grown food may be one of them. Organic food computed as one factor, atopy vs no atopy show a significant result towards protection against atopy (OR 0.63).
<b>Confounders adjusted for</b>	Other factors of anthroposophic lifestyle are not adjusted for.
<b>Relevance for our assessment purpose</b>	Some, especially in the discussion. Antroposophic lifestyle contributes with more factors than organic food which may be important for the outcome.

<b>Reference</b>	<b>Kummeling, I., Thijs, C., Huber, M., van de Vijver, L. P., Sniijders, B. E., Penders, J., Dagnelie, P. C. (2008). Consumption of organic foods and risk of atopic disease during the first 2 years of life in the Netherlands. <i>British Journal of Nutrition</i>, 99(3), 598-605</b>
<b>Study design and type</b>	Prospective epidemiological birth cohort study of children from birth to two years of life based on parental reports and assessment of sensitisation.
<b>Objective</b>	To evaluate whether early life organic food consumption was associated with developing atopic manifestations in the first two years of life.
<b>Number of participants, country and age</b>	2834 participants, 2598 were followed up until two years. Detailed questionnaires at age 3, 7, 12 and 24 months.
<b>Baseline characteristics of study subjects/cells</b>	Birth cohort recruited through Pregnancy related Pelvic Girdle Pain study (2343), and specific alternative recruitment channels (491).
<b>Exposure, substances, food (type and amount)</b>	Organic food consumption by the mother from gestational week 34 and by the child in the second year of life.
<b>Exposure; nature of the food (type and amount)</b>	Meat, eggs, vegetables, fruit, dairy products, bread and/or dry products (pasta, rice, beans and wheat) and whether these were conventionally or organically produced.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Eczema and/or wheeze occurrence classification by questionnaire. Total IgE measured by Radio Immune Assay (RIA) in serum samples, specific IgE measured by radioallergosorbent test.
<b>Follow-up period, drop-outs</b>	Two years. Three children excluded with Down`s syndrome and sixty-seven for prematurity, resulting in 2764 participants. Loss to follow-up 93.4%, 2598 followed to two years. 2135 in the conventional and 463 in the organic sub-cohort.
<b>Statistical analysis</b>	Logistic regression. Multivariate logistic regression fitted to control for confounders. Linear regression to evaluate differences in geometric means of total IgE. Separate analyses of conventional vs alternative sub-cohort revealed no differences, thus all infants were combined in the final models.
<b>Results</b>	Trend for association between organic food consumption and reduced risk of eczema, but not statistically significant. There was an association between consuming strictly organic dairy products and reduced risk of eczema. The proportion of organic food within the total diet was neither associated with eczema, nor with recurrent or prolonged wheeze, and no relation with atopic sensitisation.
<b>Conclusion</b>	The consumption of organic dairy products within the context of an organic diet in pregnant and lactating mothers is associated with reduced risk of eczema.
<b>Confounders adjusted for</b>	Sex, maternal education, BMI at age 1 year, parental history of allergy, sibling history, breastfeeding, day-care attendance, pets, smoking i environment, antibiotic intake, vegetarian diet.
<b>Relevance for our assessment purpose</b>	Yes.

<b>Reference</b>	<b>Smit, L. A., Zuurbier, M., Doekes, G., Wouters, I. M., Heederik, D., &amp; Douwes, J. (2007). Hay fever and asthma symptoms in conventional and organic farmers in The Netherlands. <i>Occupational and Environmental Medicine</i>, 64(2), 101-107</b>
<b>Study design and type</b>	Questionnaire survey in adult conventional and organic farmers.
<b>Objective</b>	To compare the prevalence of respiratory symptoms among adult organic and conventional farmers. Symptoms in farmers compared with a general non-farming population. Furthermore to what extent farming exposures during childhood combined with current farming practice affect the prevalence of asthma-like symptoms and hay fever in organic and conventional farmers.
<b>Number of participants, country and age</b>	1013 organic and 1846 conventional farmers were tried recruited. Participating 593 ORG 1205 CV 2679 subjects from the Dutch part of the European Community Respiratory Health Survey.
<b>Baseline characteristics of study subjects/cells</b>	CV male 658 (54.6%), ORG 317 (53.5%). Mean age CV45.5, ORG 44.2. Years farm work 21.8 and 16.6. Education level low or medium CV 658 (80.8%), ORG 325 (55.5%). High CV 229 (19.2%), ORG 261 (44.5%). Asthma/hay fever in parents or sibling CV 16.1%/17.1%, ORG 16.3%/ 22%. Smoking history.
<b>Exposure, substances, food (type and amount)</b>	Organic vs conventional farming - livestock and crop.
<b>Exposure; nature of the food (type and amount)</b>	
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Asthma-like symptoms (wheezing, shortness of breath, waking up due to shortness of breath. Hay fever symptoms. All symptoms self-reported.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	Logistic regression analyses used to compare prevalence of respiratory symptoms between farmers and non-farmers. Farming characteristics in association with asthma and hay fever in univariate logistic regression model for CV and ORG farmers separately, and for all farmers together.
<b>Results</b>	Farmers reported significantly less often asthma or asthma-like symptoms than non-farmers. Almost all asthma symptoms were less prevalent in ORG farmers than CV farmers. Significant for waking up and wheezing with shortness of breath. Hay fever was reported more commonly among ORG farmers than CV farmers. The association ORG farming and hay fever decreased after adjusting for confounders in a multiplke logistic regression model. The same model confirmed that currently keeping livestock and growing up on a farm were associated with a two times lower prevalence of hay fever.
<b>Conclusion</b>	Wheezing with shortness of breath was less prevalent in ORG than in CV farmers, suggesting a lower risk of asthma-like symptoms in organic farmers. The prevalence of hay fever is decreased in livestock farmers (ORG and CV) who have lived on a farm during childhood compared with crop farmers without a farm childhood.
<b>Confounders adjusted for</b>	Age, sex, smoking habits.
<b>Relevance for our assessment purpose</b>	Yes, well performed human study.

<b>Reference</b>	<b>Abell, A., Ernst, E., &amp; Bonde, J. P. (1994). High sperm density among members of organic farmers' association. Lancet, 343(8911), 1498</b>
<b>Study design and type</b>	Cross-sectional study.
<b>Objective</b>	To compare semen quality from organic farmers and workers in an organic association with workers in a conventional farmers association.
<b>Number of participants, country and age</b>	Human study from Denmark performed in 30 volunteers where 16 were organic farmers, 7 were employed in organic farmers association and 7 were linked in other ways to organic farming (OF group). At least 50% of the total dairy products were organic in 28 of this group the year before. Control groups were Flexoprints (n=19), Electricians (n=22) and Metal workers (n=32). Age between 20-50 years.
<b>Baseline characteristics of study subjects/cells</b>	The electricians were significantly younger compared with the OF. The OF had significant shorter continence period (1.3 days) compared with the other groups (3-4 days).
<b>Exposure, substances, food (type and amount)</b>	Use of organic food and at least 50% of the dairy products were organic the year before.
<b>Exposure; nature of the food (type and amount)</b>	
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Sperm count and density, by manual counting within an hour of delivery.
<b>Follow-up period, drop-outs</b>	Not available.
<b>Statistical analysis</b>	Wilcoxon rank-sum test and multivariate regression model.
<b>Results</b>	The OF had significantly higher sperm density compared with the other three groups in spite of shorter continence period. Within the group of OF there was a significant correlation between continence period and semen volume, but the volume was not correlated with sperm density. The total sperm count (volume*density) was not significantly higher in the OF group compared with the blue collar workers. When men with a shorter continence period than 24 h. were removed a total higher sperm count were seen among the OF group compared with the control groups.
<b>Conclusion</b>	Higher sperm density in OF group compared with the control groups.
<b>Confounders adjusted for</b>	Only adjusted for continence period. Not for age or other possible confounders.
<b>Relevance for our assessment purpose</b>	Yes, a study of human vitality and use of organic food items. A letter in Lancet – not a full article.

<b>Reference</b>	<b>Jensen, T. K., Giwercman, A., Carlsen, E., Scheike, T., &amp; Skakkebaek, N. E. (1996). Semen quality among members of organic food associations in Zealand, Denmark. <i>Lancet</i>, 347(9018), 1844</b>
<b>Study design and type</b>	Cross-sectional study.
<b>Objective</b>	To examine if organic farmers with at least 25% of food intake from organic food had better semen quality compared with controls.
<b>Number of participants, country and age</b>	Of 130 men with a diet consisting of at least 25% organic products 55 (42%) agreed to participate and delivered a semen sample. From 797 men working in an airline company, 141 (18%) were recruited as controls.
<b>Baseline characteristics of study subjects/cells</b>	Sexual active men. The age of the organic food users were 34.6 (4.9) years and the age of the controls were 33.3 (6.1).
<b>Exposure, substances, food (type and amount)</b>	At least 25% organic foods vs. all conventional food.
<b>Exposure; nature of the food (type and amount)</b>	
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Number of sperm cell counted by Bürker-Türk chamber.
<b>Follow-up period, drop-outs</b>	Not available.
<b>Statistical analysis</b>	Multiple linear regression of logarithmic transformed semen parameters controlled for age, length of abstinence and season at delivery.
<b>Results</b>	Sperm concentration was 43% (3.2-98.9%) higher among the organic food users compared with controls. Sperm volume, total sperm count and sperm morphology was not statistically different. No clear dose-response association between eating habits and semen quality was found.
<b>Conclusion</b>	The authors suggested that general lifestyle and /or geographic factors may have had effect on the sperm concentration.
<b>Confounders adjusted for</b>	Age, length of abstinence and season at delivery.
<b>Relevance for our assessment purpose</b>	Yes, the study is relevant, but is reported as a letter to Lancet.

<b>Reference</b>	<b>Juhler, R. K., Larsen, S. B., Meyer, O., Jensen, N. D., Spano, M., Giwercman, A., &amp; Bonde, J. P. (1999). Human semen quality in relation to dietary pesticide exposure and organic diet. Archives of Environmental Contamination and Toxicology, 37(3), 415-423</b>
<b>Study design and type</b>	Cross-sectional study.
<b>Objective</b>	To examine if organic farmers had higher semen quality compared with conventional farmers.
<b>Number of participants, country and age</b>	171 conventional farmers and 85 organic farmers in Denmark. Overall participation rate was 32%.
<b>Baseline characteristics of study subjects/cells</b>	FFQ and one semen specimen were collected from each participant before the spraying season in 1995/1996.
<b>Exposure, substances, food (type and amount)</b>	The participants were divided into three groups; one with no organic intake (N), one group consisting of farmers with intake of 1-49% organic (M) and one with high intake 50-100% organic food intake (H).
<b>Exposure; nature of the food (type and amount)</b>	Vegetables, fruit, but also meat, milk and bread. The intake of 40 different pesticides were estimated from FFQ, general portion sizes and pesticides concentration in food commodities as obtained from the National Danish Food Monitoring Program.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Pesticide intake and sperm quality.
<b>Follow-up period, drop-outs</b>	Not available.
<b>Statistical analysis</b>	Kruskal-Wallis test. Multiple linear regressions were used to compare semen quality and reproductive hormone measurements between N and H group.
<b>Results</b>	Organic farmers had significantly lower exposure to pesticides but the dietary intake of pesticides was below 1% of ADI in all groups except for dithiocarbamates (2.2% of ADI) and 2-phenylphenol (1.1% of ADI) in the N group. The N-group had a significant lower proportion of morphologically normal spermatozoa, but 14 other semen parameters were not significant different between the two groups.
<b>Conclusion</b>	There was a significant difference in intake of five pesticides between the H and N group. The estimated intake of 40 pesticides did not entail a risk of impaired semen quality.
<b>Confounders adjusted for</b>	Potential confounders were selected by their biological relevance. Age, semen spillage, sexual abstinence, fever previous 3 months, smoking, alcohol intake and self-reported reproductive disease.
<b>Relevance for our assessment purpose</b>	Yes, the study is relevant.

<b>Reference</b>	Larsen, S. B., Spano, M., Giwercman, A., & Bonde, J. P. (1999). Semen quality and sex hormones among organic and traditional Danish farmers. ASCLEPIOS Study Group. <i>Occupational and Environmental Medicine</i> , 56(2), 139-144
<b>Study design and type</b>	Cross-sectional study.
<b>Objective</b>	To examine if organic farmers had higher sperm concentration than conventional farmers.
<b>Number of participants, country and age</b>	171 conventional farmers and 85 organic farmers in Denmark (same study as Juhler).
<b>Baseline characteristics of study subjects/cells</b>	FFQ and one semen specimen were collected from each participant before the spraying season.
<b>Exposure, substances, food (type and amount)</b>	The participants were divided into organic farmers and conventional farmers (two groups).
<b>Exposure; nature of the food (type and amount)</b>	Vegetables, fruit, but also meat, milk and bread. The intake was estimated with use of a FFQ, general portion sizes.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Semen quality and sex hormones.
<b>Follow-up period, drop-outs</b>	Not available.
<b>Statistical analysis</b>	Multiple linear regressions were used to compare semen quality. Most variables were log-transformed.
<b>Results</b>	The median sperm concentration from conventional and organic farmers was 58 vs. 64 million/ml respectively. After adjustment for several confounders sperm concentration, total count, proportion of non-vital spermatozoa, sperm chromatin structure and motility variables were not significantly different.
<b>Conclusion</b>	Despite slight differences in concentrations of reproductive hormones, no significant differences in conventional measures of semen quality were found between organic and conventional farmers.
<b>Confounders adjusted for</b>	Potential confounders were selected by their biological relevance. Age, semen spillage, sexual abstinence, fever previous 3 months, smoking, alcohol intake and self-reported reproductive disease.
<b>Relevance for our assessment purpose</b>	Yes, the study is relevant.

<b>Reference</b>	Christensen, J. S., Asklund, C., Skakkebaek, N. E., Jorgensen, N., Andersen, H. R., Jorgensen, T. M., Jensen, T. K. (2013). Association between organic dietary choice during pregnancy and hypospadias in offspring: a study of mothers of 306 boys operated on for hypospadias. <i>Journal of Urology</i> , 189(3), 1077-1082
<b>Study design and type</b>	A case control study performed in 306 boys with hypospadias undergoing surgery in 2004 -2005 and 306 controls.
<b>Objective</b>	To examine if maternal choice of organic food items in pregnancy was associated with hypospadias in the offspring.
<b>Number of participants, country and age</b>	306 cases and 306 controls in Denmark. Age is not mentioned but usually boys are operated between 12 and 18 months of age.
<b>Baseline characteristics of study subjects/cells</b>	Mothers were characterised, and hypospadias were correlated with mothers with low education, parity and use of assisted reproductive technology, in addition to father's reproductive disorder.
<b>Exposure, substances, food (type and amount)</b>	Use of organic food; non-milk dairy products, organic milk, eggs, meat, fruit and vegetables.
<b>Exposure; nature of the food (type and amount)</b>	Food items analysed were non-milk dairy products, organic milk, eggs, meat, fruit and vegetables and frequency was rarely/never and often/sometimes.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Hypospadias, recruited at time point for surgery.
<b>Follow-up period, drop-outs</b>	Not available.
<b>Statistical analysis</b>	Logistic regression.
<b>Results</b>	No findings with organic food, but significant higher OR for mother who rarely/never ate organic butter and cheese and used butter and cheese more than once per day.
<b>Conclusion</b>	Difficult to interpret – no effect of organic food but increased odds for hypospadias among boys where the mothers used conventional high fat dairy products.
<b>Confounders adjusted for</b>	Adjusted for was maternal age, alcohol consumption during first trimester and BMI.
<b>Relevance for our assessment purpose</b>	Yes, a study of human health and use of organic foods.

<b>Reference</b>	<b>De Lorenzo, A., Noce, A., Bigioni, M., Calabrese, V., Della Rocca, D. G., Di Daniele, N., . . . Di Renzo, L. (2010). The effects of Italian Mediterranean organic diet (IMOD) on health status. Current Pharmaceutical Design, 16(7), 814-824</b>
<b>Study design and type</b>	Clinical study with 14 days on conventional Italian Mediterranean diet (IMD) and 14 days on Italian Mediterranean organic diet (IMOD) performed in Roma region in 2008. Cross over study.
<b>Objective</b>	To explore whether consumption of IMD according to the “Nicotera Diet” based on conventional or organic foods could affect the body composition and chemical-clinical parameters in healthy men and in age matched patients with Chronic Kidney Disease (CKD) in order to decrease CVD risk factors and progression of renal disease.
<b>Number of participants, country and age</b>	130 male subjects completed the study: Don’t know how many were healthy and how many had CKD, but 100 healthy and 50 with CRD entered the study. Healthy men age 44.7 years (SD 14.0) and CRD 46.3 years (SD 6.0).
<b>Baseline characteristics of study subjects/cells</b>	Subjects were classified as non-obese if BMI<25 and fat mass% <30% all other were classified as preobese-obese.
<b>Exposure, substances, food (type and amount)</b>	14 days with IMD and afterwards 14 days with IMOD.
<b>Exposure; nature of the food (type and amount)</b>	The conventional and the organic food were analysed with ORAC to study antioxidant capacity.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Anthropometric measurements with DXA-scanner and blood analyses were performed. Physical activity was recorded. All well described.
<b>Follow-up period, drop-outs</b>	14 days.
<b>Statistical analysis</b>	Normality was checked and mean and SD was reported for normal distributed variables. Paired sample t-test and Mann-Whitney test for difference.
<b>Results</b>	The majority of participants were regarded as sedentary during the whole study period (=no change in physical activity). No differences in the two groups were measured between after the IMD period as compared with at inclusion. After the IMOD period no differences in BMI were measured in the healthy subjects but lean body mass increased significantly. After the IMOD the CKD patients reduced BMI and fat mass while lean body mass increased. Both for the healthy and the CKD patients, most of the biochemical variables improved with regard to CVD risk factors and for the renal patients the kidney variables also improved. There was a large reduction of the inflammatory markers in the healthy participants. The antioxidant capacity was significantly higher in all of the organically grown vegetables as compared with the conventional vegetables.
<b>Conclusion</b>	After 14 days with IMOD the risk of CVD was reduce and general health improved.
<b>Confounders adjusted for</b>	No, not necessary.
<b>Relevance for our assessment purpose</b>	Yes, the study is relevant.

## Animal Model Studies

<b>Reference</b>	Chhabra, R., Kolli, S., & Bauer, J. H. (2013). Organically grown food provides health benefits to <i>Drosophila melanogaster</i> . PLoS ONE [Electronic Resource], 8(1), e52988
<b>Study design and type</b>	Animal <i>in vivo</i> and <i>in vitro</i> study over 10 days on various diets – organic or conventional, and direct testing overall health parameter.
<b>Objective</b>	To test directly whether organically farmed food elicits beneficial health effects.
<b>Number of participants, country and age</b>	Life span analysis: 50x10 flies, stress resistance: 50x8, physical activity: 20 per vial. Fertility: 20 animals per vial. Starvation and Oxidative stress resistance: 50x8 flies. Quantitative PCR: 75 flies.
<b>Baseline characteristics of study subjects/cells</b>	Wild type strain Canton-S <i>Drosophila</i> .
<b>Exposure, substances, food (type and amount)</b>	Higher levels of essential nutrients in organic food (total protein, unsaturated fatty acids, increase in antioxidants).
<b>Exposure; nature of the food (type and amount)</b>	CV and ORG potatoes, raisins, bananas and soybean. Flies fed one or the other produce.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Longevity, fertility and oxidative stress resistance. Deaths were counted, eggs were counted. For oxidative stress determination flies were cultured in medium containing H2O2 and deaths determined twice daily. Furthermore mRNA-levels of three <i>Drosophila</i> insulin-like peptides were measured.
<b>Follow-up period, drop-outs</b>	Observation up to 60 days.
<b>Statistical analysis</b>	Log-rank tests for survival, two-way ANOVA for fertility, t-tests for QPCR.
<b>Results</b>	ORG potatoes, raisins and soy led to significant extended longevity compared to CV food, while for bananas the life span was equal. Flies fed extracts of any ORG produce had significantly more eggs per day. Flies raised on ORG potato or banana, but not raisins survived the oxidative stress treatment longer than the CV produce diet. Flies raised on ORG raisin and banana food had higher overall activity than the CV same food. mRNA levels of three Dros Insulin-like peptides were slightly increased when fed ORG potato and banana, but the changes are so close to the detection limit and may be too small to be significant.
<b>Conclusion</b>	The data suggest that organic food provide improved health outcomes. The exact molecular mechanisms of the observed health effects are unclear. Altered insulin-signaling, altered redox balance or xenohormesis may all play a role.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	The use of the <i>Drosophila</i> model may be useful not only in investigating potential health effects of various food sources, but also in unraveling different molecular pathways.

<b>Reference</b>	<b>Jensen, M. M., Halekoh, U., Stokes, C. R., &amp; Lauridsen, C. (2013). Effect of Maternal Intake of Organically or Conventionally Produced Feed on Oral Tolerance Development in Offspring Rats. Journal of Agricultural and Food Chemistry, 61(20), 4831-4838</b>
<b>Study design and type</b>	A blinded animal feeding experiment in two generations of rats.
<b>Objective</b>	To investigate the effect of maternal consumption of organically or conventionally produced feed on immunological biomarkers and their offspring's response to a novel dietary antigen.
<b>Number of participants, country and age</b>	54 female Wistar Hannover GALAS rats 6 months of age fed plant based diets 6 rats per 8 plant based diet and 6 rats on rat chow diet. Second generation rats n=134 weaned at 4 weeks and switched to rat chow.
<b>Baseline characteristics of study subjects/cells</b>	
<b>Exposure, substances, food (type and amount)</b>	
<b>Exposure; nature of the food (type and amount)</b>	Crops: Potatoes, winter wheat, spring barley and fava beans supplemented with vitamin/mineral/amino acid mix. Organically grown crops were cultivated with green manure without pesticides; the conventional with inorganic fertilisers and with pesticides. Control group on rat chow.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	General health: body weight and relative spleen weight. Immunological biomarkers: Immunoglobulins in plasma and milk. T cell analysis by flow cytometry, ELISA for cytokines, TNF-alfa and PGE2 in isolated spleen cells and blood. Subpopulations ratio and T/B/NK cell distribution in blood, spleen, Peyer's Plaques. Oral tolerance induction and quantification of OVA-specific IgG, IgG1 and IgG2a.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	The responses analysed as a randomised block experiment with repeated measurements where the four blocks were defined by the levels of year crossed with the levels of location.
<b>Results</b>	No differences in the effects of maternal ORG or CV diet during pregnancy or lactation on selected immunological parameters in the offspring. No difference in OVA responses in the rats fed ORG or CV diet; however a marked difference between plant fed rats and the control group fed chow.
<b>Conclusion</b>	No differences on immune parameters or induction of oral tolerance between rats fed ORG or CV plant diet. Oral tolerance development is influenced by the mother's diet as the chow diet fed rats developed tolerance and the plant-based diets (ORG or CV) did not.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Yes, as a model for human studies of tolerance induction.

<b>Reference</b>	<b>Srednicka-Tober, D., Baranski, M., Gromadzka-Ostrowska, J., Skwarlo-Sonta, K., Rembialkowska, E., Hajslova, J., Leifert, C. (2013). Effect of Crop Protection and Fertilization Regimes Used in Organic and Conventional Production Systems on Feed Composition and Physiological Parameters in Rats. Journal of Agricultural and Food Chemistry, 61(5), 1017-1029</b>
<b>Study design and type</b>	Rat feeding study with crops from 4 different cultivation systems: The study was designed to obtain data for formulation of hypothesis for future dietary intervention studies.
<b>Objective</b>	To quantify the effects of different cultivation regimes on composition of rat feeds, body composition, growth, hormonal and immune status parameters.
<b>Number of participants, country and age</b>	32 female 16 male Wistar rats, all female rats became pregnant and gave birth. 16 dietary groups with one male and two females pro group. At three weeks two male pups from each group placed in individual cages and maintained on the same diet as their parents for another 9 weeks.
<b>Baseline characteristics of study subjects/cells</b>	
<b>Exposure, substances, food (type and amount)</b>	Four different cultivation systems: 1)Organic fertilisation and crop protection, 2)Organic fertilisation and conventional crop protection, 3) conventional fertilisation and organic crop protection, 4) conventional fertilisation and crop protection. Feeding each regime in pregnant and lactating mother rats and offspring from weaning until termination after 9 weeks.
<b>Exposure; nature of the food (type and amount)</b>	The experimental crops were wheat, potato, carrots and onion supplemented with lactalbumin, casein, rapeseed oil, minerals and vitamins.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Body composition, weight, haematological parameters. Insulin and leptin plasma concentrations .measured with radio immune assay as were Insuline-like-growth-factor-1corticosterone and testosterone plasma concentrations. IgA measured by ELISA in plasma. Cells isolated from spleen were cultured for analyses of spontaneous and mitogen stimulated proliferation.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	The main effects of interaction of crop protection and fertilisation management were assayed using analyses of variance derived from linear mixed-effects levels. Differences between the four crop management combinations were tested using Tukey's honestly significant difference test.
<b>Results</b>	Growth rate similar for all feeds, and final body weight not significantly affected by treatment. No effect on body fat. Organic fertilisation and crop protection resulted in higher concentrations of leptin, and IGF-1. Organic fertilisation with or without organic crop protection resulted in higher corticosteron and lower concentration of testosterone. Lipopolysaccharid stimulated lymphocyte proliferation was low with no stimulation of cells from rats on organically fertilised crops. Feeds from organically fertilisation and organic crop protection resulted in an increase in spontaneous proliferation, while organic fertilisation and conventional crop protection led to a decrease in Con-A stimulated proliferation. No significant effect on IgA or CRP by crop management regimes.
<b>Conclusion</b>	Crop production practices (fertilisation and crop protection) can modulate the composition of feeds and thus influence animal physiology, especially immune status.
<b>Confounders adjusted for</b>	Not adjusted for different bioavailability of secondary metabolites in the feeds, storage decrease of compounds like vitamins and minerals.
<b>Relevance for our assessment purpose</b>	Yes, well performed and high number of animals used.

<b>Reference</b>	<b>Jensen, M. M., Jorgensen, H., Halekoh, U., Watzl, B., Thorup-Kristensen, K., &amp; Lauridsen, C. (2012). Health biomarkers in a rat model after intake of organically grown carrots. Journal of the Science of Food and Agriculture, 92(15), 2936-2943</b>
<b>Study design and type</b>	Animal <i>in vivo</i> and <i>in vitro</i> study two consecutive years; year 1: 95 days, year 2: 63 days.
<b>Objective</b>	To investigate if consumption of carrots as whole food, grown under different cultivation systems during two consecutive years could have an impact on health.
<b>Number of participants, country and age</b>	50 rats per year were evaluated. GK/MO/Tac rats.
<b>Baseline characteristics of study subjects/cells</b>	Inbred strain developing type 2 diabetes characteristics.
<b>Exposure, substances, food (type and amount)</b>	Protein, nitrate, dietary fibre.
<b>Exposure; nature of the food (type and amount)</b>	Carrots grown conventional, O1: minimalistic organic, O2: organic cultivation system with intensive use of cover crops, and O3: very organic cultivation using green manure, cover crops and intercrops.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Digestibility, body and organ weight, antioxidant status, immunological response.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	
<b>Results</b>	Year no effect on chemical composition of carrots, no difference in dietary fibre, no difference between carrots grown conventional or organic in fibre or nutrients. General trend towards higher levels of all measured plasma nutrients in a normal diet compared with carrot-based diets, not significant. Plasma alfa-tocopherol in rats fed O2 than C in year 2, no effect in year 1. No other differences in antioxidants. No difference in immunological responses.
<b>Conclusion</b>	Little impact of year or cultivation system on carrot quality and little or no effect of cultivation system on biomarkers of health and immunity.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Yes as a model for human studies.

<b>Reference</b>	<b>Roselli, M., Finamore, A., Brasili, E., Capuani, G., Kristensen, H. L., Micheloni, C., &amp; Mengheri, E. (2012). Impact of organic and conventional carrots on intestinal and peripheral immunity. Journal of the Science of Food and Agriculture, 92(14), 2913-2922</b>
<b>Study design and type</b>	<i>In vitro</i> study of systemic and intestinal immunity in mice fed organic and conventional carrots. The study comprises two production years and three varieties grown in two different geographic locations.
<b>Objective</b>	To provide novel information on the impact of ORG carrots on health.
<b>Number of participants, country and age</b>	Balb-c mice 21 days old fed <i>ad libitum</i> for 30 days on ORG (3 different ORG preparations) or CV carrot-containing diets. Diet without carrots as control. At least 8 mice for each group of diets.
<b>Baseline characteristics of study subjects/cells</b>	Lymphocytes from spleens washed and suspended in PBS; intraepithelial lymphocytes (IEL) and lamina propria lymphocytes (LPL) separated from colon.
<b>Exposure, substances, food (type and amount)</b>	
<b>Exposure; nature of the food (type and amount)</b>	Carrots cultivated in 2-year field trials in Denmark and Italy. In Denmark four different cropping systems for vegetable production: One CV and three ORG. The three ORG crop rotations had increasing content of fertility-building crops for nutrient management and natural mechanisms for pest regulation. The Italian field trials performed with two different carrot varieties and laid out at two farms, one CV and one ORG. The mice were fed <i>ad lib</i> .
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Body weight, lymphocyte population percentages of IELs, LPLs blood and spleen measured by flow cytometry. Pro and anti-inflammatory cytokines were measured in serum.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	Significance of differences were evaluated by one-way analysis of variance (ANOVA), followed by Fisher`s test – p-values <0.05 considered significant.
<b>Results</b>	Year effect stronger than cultivation system (ORG vs CV) within the same year. ORG carrots induced an immune stimulation and influenced some immune phenotypes; an expansion of T-reg cells (crucial for the maintenance of immune homeostasis). The cytokine secretion study shows that ORG carrots did not induce an inflammatory status in mice.
<b>Conclusion</b>	The study indicates an immune stimulation by ORG carrots, especially the “more organic” two groups of diet. Both intestinal and peripheral immunity including an expansion of T-reg cells. The concept of safety of ORG products were reinforced (absence of possible contaminants) shown by the cytokine results.
<b>Confounders adjusted for</b>	Weather conditions and year of growth are confounders discussed.
<b>Relevance for our assessment purpose</b>	Yes as a model for human studies.

<b>Reference</b>	<b>Skwarlo-Sonta, K., Rembialkowska, E., Gromadzka-Ostrowska, J., Srednicka-Tober, D., Baranski, M., Krolikowski, T., Leifert, C. (2011). Response of animal physiology to organic versus conventional food production methods. <i>Njas-Wageningen Journal of Life Sciences</i>, 58(3-4), 89-96</b>
<b>Study design and type</b>	A dietary intervention study in rats fed crops grown under conventional or organic conditions during two consecutive growing seasons.
<b>Objective</b>	To compare the effects of crops produced in agricultural systems with contrasting combinations of fertilisation and crop protection regimes on physiological, hormonal and immune system parameters in rats.
<b>Number of participants, country and age</b>	96 rats per year, one generation rats in the first year and two generations in the second year. 6 rats per group in 16 feeding groups.
<b>Baseline characteristics of study subjects/cells</b>	Parental male and female rats randomly assigned to the 16 feeding groups and fed on experimental diet for 3 weeks. Pregnant rats were fed on their respective feeds until delivery, followed by 3 weeks suckling. Pups were weaned and male pups maintained on the same feed as mothers for another 9 weeks. Feed and water supply <i>ad lib</i> .
<b>Exposure, substances, food (type and amount)</b>	Crop protection based on conventional farming standards vs crop protection regimes permitted under organic farming standards inorganic fertilisers vs manure-compost.
<b>Exposure; nature of the food (type and amount)</b>	Four crops (wheat/barley, potato, carrot, onion) produced under four different production systems: 1)organic, 2)low input 1 (combining crop protection regime for organic with the inorganic fertilisation regime in the conventional system) 3)low input 2 (combining pesticide-based crop protection with manure-compost fertilisation regime) 4)conventional.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Growth dynamics, body composition, haematological parameters (RBC, PCV, Hb; WMC), hormonal status (insulin, growth hormone, IGF-1, corticosteron, testosterone) antioxidant capacity of plasma (trolox equivalent antioxidant capacity (plasma TEAC)) , immune status (IgA and IgG, splenocyte proliferation spontaneous and mitogen-stimulated).
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	3-factor ANOVA with crop protection, fertility management and year or rat generation as main factors and replicate blocks as random variables. Multivariate analyses.
<b>Results</b>	Polyphenol and lutein higher in ORG than CV crops, protein content higher in CV. Organic fertilisation greater impact on polyphenol than organic crop protection. More differences between years grown than between regimes. Growth higher in rats fed on CV fertilised crops. No differences in haematological parameters between ORG and CV, but higher Hb in both fully ORG and fully CV than in the two low input crops. Higher plasma TEAC values in rats fed on ORG, no association between feed TEAC and plasma TEAC.
<b>Conclusion</b>	The production method affected the composition of feed (macronutrients and bioactive compounds), rat growth body composition and hormonal balance. However, the differences in feed composition could not be directly linked to the observed physiological differences. Important question: whether and to what extent is observed physiological effects beneficial or harmful.
<b>Confounders adjusted for</b>	Age when separated from mothers (not adjusted for in this study).
<b>Relevance for our assessment purpose</b>	Yes as a model for human studies.

<b>Reference</b>	<b>Huber, M., Van De Vijver, L. P. L., Parmentier, H., Savelkoul, H., Coulier, L., Wopereis, S., Hoogenboom, R. A. P. (2010). Effects of organically and conventionally produced feed on biomarkers of health in a chicken model. <i>British Journal of Nutrition</i>, 103(5), 663-676</b>
<b>Study design and type</b>	A blinded animal feeding experiment in two generations of chicken. The second generation received experimental diets from 11 – 13.3 weeks of age.
<b>Objective</b>	To identify biomarkers of the effect of organic feed on health in chickens, focusing on immune response as well as specific compartment, growth and metabolism.
<b>Number of participants, country and age</b>	72 first generation and 150 second generation.
<b>Baseline characteristics of study subjects/cells</b>	Chickens from Wageningen Selection lines selected for primary high or low antibody response to sheep red blood cells immunisation at 35 days of age. A randomly bred control group was included.
<b>Exposure, substances, food (type and amount)</b>	Amount of protein, several amino acids, fat and carbohydrate. LPS endotoxins, gram negative bacteria.
<b>Exposure; nature of the food (type and amount)</b>	Six ingredients in the feeds: Wheat, barley, triticale, peas, maize, soya produced either organically or conventionally. Supplemented with amino acids and potato protein as well as a mix of vitamins and minerals. Animals were allowed free feeding and water supply.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	General health parameters, weight and feed intake, eggs produced. Parameters of infection, innate and specific immunity, humoral and cellular. Immune response to stimulation with KLH.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	Univariate statistics computed for measurements on chickens grouped by organic and conventional feed. Mean differences tested with ANOVA and Wilcoxon.
<b>Results</b>	No difference in body weight (except for one animal line fed conventional grown food who were significantly heavier), growth rate or number, weight or quality of the eggs. Differences in immune parameters and immune responses.
<b>Conclusion</b>	Enhanced immune reactivity, stronger reaction to immune challenge and slightly stronger catch up growth after the challenge on organic feed. From the present data is concluded that diets from different production systems, organic or conventional can induce physiological changes in two generations of chickens.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	The study was designed to identify biomarkers of health in chickens, but relevant as the model is useful for identifying markers further human studies.

<b>Reference</b>	Lauridsen, C., Yong, C., Halekoh, U., Bugel, S. H., Brandt, K., Christensen, L. P., & Jorgensen, H. (2008). Rats show differences in some biomarkers of health when eating diets based on ingredients produced with three different cultivation strategies. <i>Journal of the Science of Food and Agriculture</i> , 88(4), 720-732
<b>Study design and type</b>	Animal feeding study based with <i>in vivo</i> and post mortem parts. Experimental diets provided from one month before the parents started breeding and until the end of the study when two-generation rats were 44 weeks old.
<b>Objective</b>	To define which measurable aspects of health (if any) would be most affected through differences in production methods.
<b>Number of participants, country and age</b>	36 female GK/mol rats after weaning of their first litter (age 19 weeks). Denmark.
<b>Baseline characteristics of study subjects/cells</b>	Rats were housed three per cage of a given experimental diet. Water and feed provided <i>ad lib</i> .
<b>Exposure, substances, food (type and amount)</b>	Three experimental diets from three different cultivation systems: LIminusP: Low input of nutrients through plant residues and animal manure, using similar or lower levels of organic inputs than standard organic practice for the crop and no pesticides. LIplusP: similarly low input of nutrients supplemented with as much pesticides as allowed. HIplusP: High input of nutrients through mineral fertilisers and as much pesticides as allowed corresponding to standard conventional practice for the crop.
<b>Exposure; nature of the food (type and amount)</b>	The three diets were formulated to meet or exceed National Research Council recommendations for growing and reproducing rats. The diets were composed of potatoes, carrots, apples, peas, green kale and rapeseed oil.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Protein and energy digestability through quantitative collection of faeces and urine. Heat production calculated from gas exchange measurements. Gastric emptying and liver metabolic function assayed in a breath test. Biochemical measurements of blood and tissues: Fatty acid profiles of liver, plasma and adipose tissue. Serum concentrations of total protein, glucose, urea-N, free fatty acids and aspartate aminotransferase measured in auto-analyser, Vitamins (tocopherol, vitamin A and beta-carotene in plasma, adipose tissue and liver analysed by HPLC. Serum concentrations of total, HDL and LDL cholesterol measured enzymatically, IgG, IgA and IgM measured in ELISA kits.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	Results of the statistical analyses given as least square means and their standard errors. Fitting for the models generated using the mixed procedure of SAS/STAT software, version 8.2.
<b>Results</b>	LIplusP diet higher proportion of oleic acid and lower proportion of linoleic acid than the other diets. The rapeseed oil of LIplusP contained less vitamin E. No detectable pesticide residues in any diet. No important differences in nutrient digestibility or energy metabolism. The LIminusP rats had lower content of adipose tissue compared to HIplusP rats. Lower physical energy of LIminusP rats during the day, no differences during the night. No differences of vitamin E in adipose tissue by dietary treatment although differences in plasma. Significantly elevated IgG concentration in LIminusP and LIplusP rats.
<b>Conclusion</b>	Even though the diets were nutritionally similar some differences (alfa-tocopherol, IgG, daytime activity, volume of adipose tissue) appeared which should be taken into consideration in future studies.
<b>Confounders adjusted for</b>	The production systems used were not replicated and represented combinations of relevant methods rather than actual commercial systems.
<b>Relevance for our assessment purpose</b>	Yes – rats used were from a strain which can develop diet-related type 2 diabetes, which is relevant for European human population.

<b>Reference</b>	<b>Finamore, A., Britti, M. S., Roselli, M., Belovino, D., Gaetani, S., &amp; Mengheri, E. (2004). Novel approach for food safety evaluation. Results of a pilot experiment to evaluate organic and conventional foods. Journal of Agricultural and Food Chemistry, 52(24), 7425-7431</b>
<b>Study design and type</b>	<i>In vitro</i> animal study 30 days feeding.
<b>Objective</b>	A novel approach to evaluate the potential health risk induced by long-term consumption of organic vs conventional cereals, i.e. the assay of sensitive markers of cell function in vulnerable conditions.
<b>Number of participants, country and age</b>	Male Sprague-Dawley rats at weaning. Results based on ten rats per group.
<b>Baseline characteristics of study subjects/cells</b>	Well-nourished vs protein-energy mal nourished rats fed organic vs conventional cereals. Mesenteric lymph nodes and spleen cells – peripheral blood were collected at day 30.
<b>Exposure, substances, food (type and amount)</b>	Fusarium, penicillium, Aspergillus and mycotoxins in organic vs conventional wheat.
<b>Exposure; nature of the food (type and amount)</b>	
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Body weight, lymphocyte proliferation (intestinal and peripheral sites) and liver acute phase reaction.
<b>Follow-up period, drop-outs</b>	
<b>Statistical analysis</b>	One way ANOVA followed by Fisher test. Differences with $p < 0.5$ significant.
<b>Results</b>	Proliferative capacity of lymphocytes from WN-Org rats similar to WN-CV rats. In PEM rats org wheat did not affect the proliferative capacity. Lymphocytes of PEM rats cultured with rat serum (instead of fetal calf serum), the mitogen stimulation was lower in PEM-CV than PEM-ORG suggesting contaminants present in conventional and not organic wheat.
<b>Conclusion</b>	No higher risk of introducing toxic compounds with organic compared to conventional wheat that could affect fundamental cell functions. Functional assay may reveal contaminants present which is not shown with chemical assays alone.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Focus on more extreme conditions (mal-nourishment), interesting as a model for future human studies.

## Biomarker Studies

Reference	Soltoft, M., Bysted, A., Madsen, K. H., Mark, A. B., Bugel, S. G., Nielsen, J., & Knuthsen, P. (2011). Effects of organic and conventional growth systems on the content of carotenoids in carrot roots, and on intake and plasma status of carotenoids in humans. <i>Journal of the Science of Food and Agriculture</i> , 91(4), 767-775
<b>Study design and type</b>	I: Crops were grown conventional (C) or organic using animal manure (OA) or using cover crops (OB). II: A double-blinded, crossover human study for two consecutive years.
<b>Objective</b>	I: To study the effect of organic versus conventional growth system on the content of carotenoids in carrots. II: To measure the concentration of carotenoid in plasma from subjects who consumed organic or conventional diets.
<b>Number of participants, country and age</b>	N=18, Denmark, age 25.1±6.7 and 26.0±7.1 and BMI 21.8±1.6 and 24.0±2.3.
<b>Baseline characteristics of study subjects/cells</b>	Adult male subjects, non-smokers, without medication use, two different groups from Copenhagen.
<b>Exposure, substances, food (type and amount)</b>	6 subjects consumed diet C, 6 subjects OA and 6 subjects consumed diet OB.
<b>Exposure; nature of the food (type and amount)</b>	Diets C, OA or OB. Intervention 3 x 12 days, wash out period 2 weeks, two replicates of each diet, total six diets.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	I: Content of carotenoids in carrots cultivated by three systems. II: Carotenoids concentration in plasma after consumption of foods produced by three systems.
<b>Follow-up period, drop-outs</b>	12 days.
<b>Statistical analysis</b>	ANOVA, t-test, post-hoc tests.
<b>Results</b>	The concentration of $\beta$ -carotene increased in plasma after the intervention period, Group C, 0.30±0.19 to 0.69±0.29 $\mu\text{g/ml}$ ( $p<0.05$ ), group OA, from 0.26±0.17 to 0.49±0.22 $\mu\text{g/ml}$ ( $p<0.05$ ) and group OB, from 0.34±0.55 to 0.55±0.20 $\mu\text{g/ml}$ ( $p<0.05$ ). Similar results were obtained after the dietary intervention trial in the second year. However, the consumption of carrots cultivated either conventionally or organically had no significant impact on the concentration of plasma carotenoids in healthy subjects.
<b>Conclusion</b>	Carrots produced by organic or conventional growing systems had no effect on the content of carotenoids. Consumption of carrots produced by organic or conventional growing systems had no effect on plasma concentration of carotenoids in healthy individuals.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	<b>Burns-Whitmore, B. L., Haddad, E. H., Sabate, J., Jaceldo-Siegl, K., Tanzman, J., &amp; Rajaram, S. (2010). Effect of n-3 fatty acid enriched eggs and organic eggs on serum lutein in free-living lacto-ovo vegetarians. <i>European Journal of Clinical Nutrition</i>, 64(11), 1332-1337</b>
<b>Study design and type</b>	Human, randomised crossover trial of 8 weeks duration.
<b>Objective</b>	Determine lutein, zeaxanthin and beta-carotene upon eating either organic eggs or eggs enriched in n-3 fatty acids.
<b>Number of participants, country and age</b>	20, USA, age range 21-90.
<b>Baseline characteristics of study subjects/cells</b>	Healthy non-smoking lacto-ovo-vegetarians (for 3 months or more).
<b>Exposure, substances, food (type and amount)</b>	Eggs.
<b>Exposure; nature of the food (type and amount)</b>	The subjects ate 6 eggs/week.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Plasma antioxidant level.
<b>Follow-up period, drop-outs</b>	Baseline, after 8 weeks on the two egg forms with a 4 week washout in-between.
<b>Statistical analysis</b>	Log-transformed data to achieve a normal distribution. Mixed linear model and ANOVA.
<b>Results</b>	Only serum lutein increased (about 2.5-fold; $p < 0.009$ ), and then after consumption of both egg forms.
<b>Conclusion</b>	Consumption of organic eggs may increase plasma lutein.
<b>Confounders adjusted for</b>	Age, BMI, cholesterol.
<b>Relevance for our assessment purpose</b>	Yes, lutein may reduce risk of age-related macula degeneration (an eye-disorder).

<b>Reference</b>	<b>Stracke, B. A., Rufer, C. E., Bub, A., Briviba, K., Seifert, S., Kunz, C., &amp; Watzl, B. (2009). Bioavailability and nutritional effects of carotenoids from organically and conventionally produced carrots in healthy men. <i>British Journal of Nutrition</i>, 101(11), 1664-1672</b>
<b>Study design and type</b>	I: Carrots were produced by conventional or organic system. II: A double-blinded, randomised intervention study. 2 weeks.
<b>Objective</b>	To compare the content of carotenoids in carrots produced by different systems, and to study the effect of carrot consumption on antioxidant capacity, DNA strand breaks, and immune parameters in healthy subjects.
<b>Number of participants, country and age</b>	N=36, Germany, age 19-54 years.
<b>Baseline characteristics of study subjects/cells</b>	Healthy men, non-smokers, no medications.
<b>Exposure, substances, food (type and amount)</b>	Three groups (n=12/ group) consumed diets containing 200 g blanched carrots from organically or conventionally produced system. The third, control group did not consume carrots rich diets.
<b>Exposure; nature of the food (type and amount)</b>	200 g carrots/meal or no carrots/meal.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Status of markers of antioxidant system in plasma.
<b>Follow-up period, drop-outs</b>	Two weeks, washout period two weeks.
<b>Statistical analysis</b>	Unpaired t-test, repeated measures of ANOVA, Tukey-Kramer post-hoc test.
<b>Results</b>	The content of carotenoids in carrots grown organically or conventionally was not significantly different ( $121 \pm 7$ versus $116 \pm 13$ $\mu\text{g/g}$ , ( $p > 0.05$ )). The lipophilic antioxidant activity was not significantly different in the carrots produced organically or conventionally, $0.43 \pm 0.08$ versus $0.032 \pm 0.07$ , $\mu\text{mol Trolox eq/g fresh wt}$ ( $p > 0.05$ ). The concentration of $\alpha$ -carotene and $\beta$ -carotene increased significantly in plasma in organic group as well as in conventional diet group ( $p < 0.001$ , ANOVA). However, there was not a significant difference in the concentration of $\alpha$ -carotene and $\beta$ -carotene when compared organic diet and conventional diet groups ( $p > 0.05$ ). The organic or conventional diet had no significant effect on the parameters of antioxidant status after the intervention, ferric-reducing ability in plasma (FRAP) in the organic diet group, the baseline $925 \pm 15$ after the intervention $906 \pm 135$ $\text{Fe}^{2+}/\text{L}$ $\mu\text{mol/L}$ , in the conventional diet group, the baseline $921 \pm 160$ after the intervention $908 \pm 130$ and in the control group, the baseline $968 \pm 173$ after the intervention $903 \pm 135$ , ( $p > 0.05$ repeated measures ANOVA). Similarly, no significant difference was detected between the groups after the dietary intervention in TEAC, DNA strand breaks and carotenoid concentration in the peripheral blood mononuclear cells ( $p > 0.05$ ), repeated measures ANOVA).
<b>Conclusion</b>	Organically or conventionally produced carrots had no effect on the content of carotenoids. Consumption of carrots produced by the two systems had no impact on markers of antioxidant or immune system.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	<b>Briviba, K., Stracke, B. A., Rufer, C. E., Watzl, B., Weibel, F. P., &amp; Bub, A. (2007). Effect of consumption of organically and conventionally produced apples on antioxidant activity and DNA damage in humans. Journal of Agricultural and Food Chemistry, 55(19), 7716-7721</b>
<b>Study design and type</b>	A double-blind randomised, cross over study, wash-out period one week.
<b>Objective</b>	To compare the markers of plasma antioxidant status in healthy subjects consuming organically or conventionally produced apples.
<b>Number of participants, country and age</b>	6, Switzerland, 27±3 years.
<b>Baseline characteristics of study subjects/cells</b>	Non-smoking male subjects, BMI 23±3.
<b>Exposure, substances, food (type and amount)</b>	Apples.
<b>Exposure; nature of the food (type and amount)</b>	1000 g organically or conventionally produced apples.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Levels of antioxidants in plasma.
<b>Follow-up period, drop-outs</b>	24 hours.
<b>Statistical analysis</b>	Unpaired t-test, Repeated measures of ANOVA, Dunnett's post-hoc test.
<b>Results</b>	The concentration of chlorogenic acid in organically or conventionally cultivated apples was not significantly different 104.2±25.5 and 96.7±10.8 µg/g fresh weight (n=8) (p>0.05). There was not a significant difference in the concentration of sum of all phenolic compounds in two apple types, 307.9±70.6 and 320.9±66.5 µg/g fresh weight (n=8), (p>0.05). The consumption of apples cultivated with organic or conventional methods had no effect on the concentration of serum glucose, serum triacylglycerol and serum uric acid in the participants. The oxidation level of low-density lipoprotein was not altered after the consumption of apples produced organically or conventionally. Parameters of DNA damage were not affected, however, a significant reduction was recorded for the level of Endo III-specific sites after consumption of organic apples, baseline 3.69±2.61, after 4.5 hours 2.97±1.19 and after 24 hours 1.10±1.20 (p<0.05, repeated measures of ANOVA, Dunnett post-hoc test) or conventional apples, baseline 4.60±1.23, after 4.5 hours 3.05±1.87 and after 24 hours 1.19±0.73 (p<0.05 repeated measures of ANOVA, Dunnett post-hoc test). The FeCl <sub>3</sub> -induced DNA strand break was reduced, an increased protective capacity, the consumption of organic apples, baseline, 9.38±2.89, after 4.5 hours, 6.92±1.18 and after 24 hours, 5.12±1.67 (p<0.05, repeated measures of ANOVA, Dunnett post-hoc test).
<b>Conclusion</b>	Polyphenols in apples may protect DNA from oxidative stress.
<b>Confounders adjusted for</b>	No
<b>Relevance for our assessment purpose</b>	Yes

Reference	<b>Di Renzo, L., Di Pierro, D., Bigioni, M., Sodi, V., Galvano, F., Cianci, R., De Lorenzo, A. (2007). Is antioxidant plasma status in humans a consequence of the antioxidant food content influence? European Review for Medical and Pharmacological Sciences, 11(3), 185-192</b>
<b>Study design and type</b>	This study has two parts: <ol style="list-style-type: none"> <li>1. To compare the antioxidant levels, by measuring the oxygen radical absorbing capacity (ORAC), in organic and conventional fruits and vegetables.</li> <li>2. To test whether consumption of organic or conventional foods by healthy subjects affect plasma ORAC levels.</li> </ol>
<b>Objective</b>	To test whether consumption of organic or conventional foods by healthy subjects affect plasma ORAC levels.
<b>Number of participants, country and age</b>	10, Italy, 30-65 years.
<b>Baseline characteristics of study subjects/cells</b>	Healthy, non-smoker subjects who were not abusing alcohol, were free from hypertension and cardiovascular disease.
<b>Exposure, substances, food (type and amount)</b>	Organic or conventional foods.
<b>Exposure; nature of the food (type and amount)</b>	14 days.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Measurement of plasma ORAC, which reflects antioxidant status.
<b>Follow-up period, drop-outs</b>	14days.
<b>Statistical analysis</b>	Mann Whitney Test.
<b>Results</b>	Most organic foods had higher antioxidant capacity than conventional foods ( $p<0.005$ ). The concentration of ORAC increased in plasma from men after consumption of organic foods for 14 days ( $p<0.005$ ).
<b>Conclusion</b>	Polyphenols in organic foods may affect antioxidant levels in healthy subjects.
<b>Confounders adjusted for</b>	No.
<b>Relevance for our assessment purpose</b>	Yes.

<b>Reference</b>	Akçay, Y. D., Yildirim, H. K., Guvenc, U., & Sozmen, E. Y. (2004). The effects of consumption of organic and nonorganic red wine on low-density lipoprotein oxidation and antioxidant capacity in humans. <i>Nutrition Research</i> , 24(7), 541-554
<b>Study design and type</b>	Intervention study for one day, wine drinking for 15 min, organic wine or nonorganic wine.
<b>Objective</b>	To test whether consumption of organic and nonorganic wines by healthy subjects increases antioxidant capacity and affects the oxidation status of low-density lipoprotein.
<b>Number of participants, country and age</b>	6 nonsmoker men and 2 nonsmoker women. Turkey. Age 24-45 years.
<b>Baseline characteristics of study subjects/cells</b>	Healthy nonsmoker subjects who were not taking any medication.
<b>Exposure, substances, food (type and amount)</b>	Organic or nonorganic wine.
<b>Exposure; nature of the food (type and amount)</b>	Wine exposure for 15 min in one day.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Blood antioxidant capacity and effect on the oxidation level of low-density lipoprotein.
<b>Follow-up period, drop-outs</b>	6 hours.
<b>Statistical analysis</b>	Multivariate analysis.
<b>Results</b>	The antioxidant capacity increased 1 and 6 hours after the consumption of organic red wine ( $p=0.03$ and $p=0.048$ ). The antioxidant capacity decreased 70% 1 hour after and increased 82% after the consumption of nonorganic wine. Organic or nonorganic wines had no effect on the levels of oxidation of LDL.
<b>Conclusion</b>	Organic or nonorganic wines had no effect on the levels of oxidation of LDL or TBARs.
<b>Confounders adjusted for</b>	No
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	Caris-Veyrat, C., Amiot, M. J., Tyssandier, V., Grasselly, D., Buret, M., Mikolajczak, M., Borel, P. (2004). Influence of organic versus conventional agricultural practice on the antioxidant microconstituent content of tomatoes and derived purees; consequences on antioxidant plasma status in humans. <i>Journal of Agricultural and Food Chemistry</i> , 52(21), 6503-6509
<b>Study design and type</b>	Human, randomised trial of 3 weeks duration.
<b>Objective</b>	1. Determine antioxidants in tomatoes cultured organic and non-organic. 2. Determine plasma-anti-oxidants of human consuming purees based on the two culture-forms of tomatoes.
<b>Number of participants, country and age</b>	10, France, mean age 25.
<b>Baseline characteristics of study subjects/cells</b>	Healthy non-smoking females with normal BMI.
<b>Exposure, substances, food (type and amount)</b>	Tomatoes (antioxidants).
<b>Exposure; nature of the food (type and amount)</b>	The subjects ate 100g/day for 3 weeks of tomato puree (for lunch or dinner).
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Plasma antioxidant level.
<b>Follow-up period, drop-outs</b>	Baseline, after 3 weeks on tomato puree, then after 3 weeks washout.
<b>Statistical analysis</b>	ANOVA.
<b>Results</b>	Higher antioxidant levels ( $p < 0.001$ ; vitamin C and polyphenols) in tomatoes grown organic vs. non-organic. No difference in plasma antioxidant ( $p > 0.05$ ; vitamin C and lycopene) levels after 3 weeks.
<b>Conclusion</b>	Albeit higher antioxidant levels in organic tomatoes, it did not translate into higher plasma levels of plasma antioxidant levels after consumption of these tomatoes in the form of puree.
<b>Confounders adjusted for</b>	None.
<b>Relevance for our assessment purpose</b>	Yes, plasma-antioxidant level as proxy for health.

<b>Reference</b>	<b>Grinder-Pedersen, L., Rasmussen, S. E., Bugel, S., Jorgensen, L. V., Dragsted, L. O., Gundersen, V., &amp; Sandstrom, B. (2003). Effect of diets based on foods from conventional versus organic production on intake and excretion of flavonoids and markers of antioxidative defense in humans. Journal of Agricultural and Food Chemistry, 51(19), 5671-5676</b>
<b>Study design and type</b>	Human, double blind, crossover intervention. Two intervention periods. Duration 22 days. Wash out period 3 weeks. Collection of urine and plasma samples before and after the intervention.
<b>Objective</b>	To measure the concentration of 5 flavonoids in urine and markers of antioxidant defence system in subjects consuming organic or conventional diets.
<b>Number of participants, country and age</b>	16, Denmark, 21-35 years.
<b>Baseline characteristics of study subjects/cells</b>	Healthy men (n=6) and women (n=10), non-smokers, non-pregnant, mean BMI 23.4 kg/m <sup>2</sup> , average energy intake 12±2.7 MJ. No heavy physical exercise for three days, no alcohol consumption for 24 hours before the intervention.
<b>Exposure, substances, food (type and amount)</b>	Organic or conventional diet.
<b>Exposure; nature of the food (type and amount)</b>	Organic or conventional diet.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Flavonoid quercetin content in the organically produced diets (OPD) was significantly higher than in conventionally produced diets (CPD) 4198±1370 versus 2632±774 µg/10MJ, median±SD (p<0.01, Mann-Whitney U test). The concentrations of urine quercetin and kaempferol were higher in individuals consuming organic diets than in individuals consuming conventional diets. However, the concentration of was not different when tested for intake of quercetin and kaempferol.
<b>Follow-up period, drop-outs</b>	22 days.
<b>Statistical analysis</b>	Wilcoxon signed rank test and Mann-Whitney U test.
<b>Results</b>	Flavonoid quercetin content in the organically produced diets (OPD) was significantly higher than in conventionally produced diets (CPD) 4198±1370 versus 2632±774 µg/10MJ, median±SD (p<0.01, Mann-Whitney U test). The TEAC was significantly higher after the consumption of CPD than OPD 1.029±0.074 versus 0.0951±0.056 mmol/L, (p<0.05). Other plasma markers of plasma antioxidant status were not changed significantly either by the consumption of CPD or OPD. Urine quercetin concentrations were significantly higher than the baseline concentrations after the consumption of CPD and OPD (quercetin concentration at baseline 6±3 versus CPD 19±2 and OPD 27±3 µg/24 hours urine, median±SEM, (p<0.05, Wilcoxon ranks scores). Similarly, the concentration of kaempferol was significantly changed after the consumption of CPD and OPD, baseline 0.0±0.3, CPD 2±1 and OPD 5±4 µg/24 hour urine (p<0.05). The consumption of OPD caused a higher urinary quercetin and kaempferol excretion than CPD (p<0.05). The authors of this study suggest that OPD diets may have an effect on the concentration of urinary flavonoids. Urine quercetin and kaempferol concentrations were higher in the individuals consuming organic diet than individuals consuming conventional diets. Protein oxidation increased and antioxidant capacity decreased after the intervention.
<b>Conclusion</b>	Organic diet is associated with increased excretion of quercetin and kaempferol in the urine.
<b>Confounders adjusted for</b>	No
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	<b>Rist, L., Mueller, A., Barthel, C., Sniijders, B., Jansen, M., Simoez-Wust, A. P., Thijs, C. (2007). Influence of organic diet on the amount of conjugated linoleic acids in breast milk of lactating women in the Netherlands. British Journal of Nutrition, 97(4), 735-743</b>
<b>Study design and type</b>	Women provided milk 1month post-partum for fatty acid analysis.
<b>Objective</b>	To measure the amount of conjugated linoleic acid isomers (CLA) and trans-vaccenic acid (TVA) in breastmilk from women consuming mostly conventional diets or mostly organic diets.
<b>Number of participants, country and age</b>	312 women, Netherlands.
<b>Baseline characteristics of study subjects/cells</b>	146 women consumed the conventional diets and 166 women consumed the organic diets.
<b>Exposure, substances, food (type and amount)</b>	Organic or conventional diets.
<b>Exposure; nature of the food (type and amount)</b>	Organic or conventional diets.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Whether consumption of organic diet versus conventional diet by lactating women has an effect on the amount of conjugated linoleic acids in the breastmilk.
<b>Follow-up period, drop-outs</b>	Breastmilk was collected one-month; post-partum.
<b>Statistical analysis</b>	Students´ s t-test and linear regression analysis.
<b>Results</b>	The concentration of rumenic acid was significantly higher in the breastmilk collected from the women who consumed mostly organic diets than in the breastmilk collected from the women on mostly conventional diet, $0.34\pm 0.1$ (>90% organic diet), $0.29\pm 0.10$ (50-90% organic diet) $0.25\pm 0.07$ (conventional diet) g/100 weight% of total fat ( $p<0.0001$ ). Similarly, TVA concentration was higher in the breastmilk collected from the women who consumed organic diet $0.59\pm 0.16$ (>90% organic) than the women who consumed conventional diet $0.48\pm 0.21$ , $p<0.001$ ). The breastmilk from the women consuming organic diet had lower concentration of $\alpha$ -linolenic acid (C18:3n-3) and total MUFA than in the breastmilk obtained from the women on the conventional diet, $0.82\pm 0.28$ and $38.57\pm 3.20$ versus $1.05\pm 0.38$ and $40.84\pm 3.08$ , g/100 weight% of total fat ( $p<0.001$ for both). Rumenic acid remained significantly higher in the breastmilk collected from the women in the organic diet group than in the milk from women in the conventional group after adjusting several variables ( $p<0.001$ ). An increase in TVA concentration was associated with an increased intake of organic foods.
<b>Conclusion</b>	Lactating women who consumed organic diets had higher concentration of rumenic acid and TVA in breastmilk than the women who consumed conventional diets.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	Mueller, A., Thijs, C., Rist, L., Simoes-Wust, A. P., Huber, M., & Steinhart, H. (2010). Trans fatty acids in human milk are an indicator of different maternal dietary sources containing trans fatty acids. <i>Lipids</i> , 45(3), 245-251
<b>Study design and type</b>	Women provided milk 1month post-partum for the fatty acid analysis.
<b>Objective</b>	To measure the amount of trans fatty acid (TFA) isomers in the breastmilk from the women consuming mostly conventional diets or mostly organic diets.
<b>Number of participants, country and age</b>	312 women, Netherlands.
<b>Baseline characteristics of study subjects/cells</b>	146 women consumed mostly conventional diets and 166 women consumed mostly organic diets.
<b>Exposure, substances, food (type and amount)</b>	Organic or conventional diets.
<b>Exposure; nature of the food (type and amount)</b>	Organic or conventional diets.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Amount of TFA isomers in the breastmilk.
<b>Follow-up period, drop-outs</b>	Breastmilk was collected one-month, post-partum.
<b>Statistical analysis</b>	Linear regression analysis, ANOVA, Tukey-Kramer test.
<b>Results</b>	No difference was detected in the concentration of TFA in the breastmilk between the groups, mostly conventional and mostly organic diets. 9(trans)18:1/11(trans)18:1-ratio and the 10(trans)18:1/11(trans)18:1-ratio were lower in the breastmilk from the women with high dairy fat intake than the women with lower dairy fat intake. Similarly, these ratios were lower in the women who consumed mostly organic meat and dairy products than the women who consumed mostly conventional diets.
<b>Conclusion</b>	Consumption of organically produced meat and dairy products by lactating women had an effect on TFA isomers in the breastmilk.
<b>Confounders adjusted for</b>	
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	Mark, A. B., Kapolna, E., Laursen, K. H., Halekoh, U., Rasmussen, S. K., Husted, S., Bugel, S. (2013). Consumption of organic diets does not affect intake and absorption of zinc and copper in men - evidence from two cross-over trials. <i>Food &amp; Function</i> , 4(3), 409-419
<b>Study design and type</b>	Human, double-blinded, cross-over trial. 3 dietary periods, duration 12 days, and wash out period 14 days.
<b>Objective</b>	Intake and absorption of zinc and copper through organic or conventional diets.
<b>Number of participants, country and age</b>	17 in 2008, age 24.3±6.3, BMI 22.2±1.5, and 16 in 2009, age 27.8±8.9, BMI, 24.1±2.8. Denmark.
<b>Baseline characteristics of study subjects/cells</b>	Healthy, non-smokers men.
<b>Exposure, substances, food (type and amount)</b>	Organic cultivating system with animal manure (OA), organic cultivating with cover crops (OB), and conventional cultivating system (C). 9 crops grown in 2 organic agriculture systems (OA and OB). On day 8, copper and zinc isotopes were given to the study participants.
<b>Exposure; nature of the food (type and amount)</b>	Organic or conventional diets.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Absorption of copper and zinc from organic or conventional diets. Faecal samples were tested for the measurement of Cu and Zn isotope.
<b>Follow-up period, drop-outs</b>	Faecal samples were collected, baseline samples, two days before the study start and after the isotope administration (on days 8 -12).
<b>Statistical analysis</b>	ANOVA, students' t-test.
<b>Results</b>	Inositol hexakisphosphate (IP) or phytic acid fractions (IP4+IP5+IP6) were similar in all diets, OA, OB or C (2.82±0.7 g/10 MJ) thus the agriculture cultivating systems had no effect on the diet components. OA, OB or C had no effect on the intake of copper or zinc (Cu, OA, 2.13±0.02, OB 2.06±0.02, C 2.07±0.02, Zn, OA 12.79±0.66, OB, 12.38±0.88, C 12.28±0.88 (p>0.05) or absorption of copper or zinc Cu, OA 41.25±9.80, OB, 31.17±11.06, C 32.70±10.15 and Zn, OA 47.50±12.52, OB, 40.60±11.07, C diets, 44.27±10.48% in the subjects. Organic or conventional diets had no effect on the intake or absorption of copper and zinc.
<b>Conclusion</b>	The trace elements copper and zinc were absorbed by the study participants. However, organic or conventional diets did not affect the intake or absorption of trace elements.
<b>Confounders adjusted for</b>	No
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	Lu, C., Toepel, K., Irish, R., Fenske, R. A., Barr, D. B., & Bravo, R. (2006). Organic diets significantly lower children's dietary exposure to organophosphorus pesticides. <i>Environmental Health Perspectives</i> , 114(2), 260-263
<b>Study design and type</b>	Human, longitudinal design. Collection of urine for 15 days.
<b>Objective</b>	To measure the concentration of organophosphorus pesticide metabolites in urine from children consuming conventional diet, organic diet and then conventional diet.
<b>Number of participants, country and age</b>	23, USA, age 3-11 years.
<b>Baseline characteristics of study subjects/cells</b>	Healthy children.
<b>Exposure, substances, food (type and amount)</b>	Children consumed conventional diets for three days, 1-3 (phase 1), day 4-8 organic diets (phase 2) and conventional diets, day 9-15 (phase 3). Exposure for 15 days.
<b>Exposure; nature of the food (type and amount)</b>	Organic or conventional.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Measurement of the concentration of pesticide metabolites malathion and chlorpyrifos in urine after the consumption of organic diets.
<b>Follow-up period, drop-outs</b>	Urine collection for 24 hours for 15 days, twice a day.
<b>Statistical analysis</b>	ANOVA and Tukey test.
<b>Results</b>	The concentration of malathion dicarboxylic acid (MDA) was significantly lower during phase 2 than phase 1 or phase 3, $0.3 \pm 0.9 \mu\text{g/L}$ versus $2.9 \pm 5.0$ and $4.4 \pm 12.4 \mu\text{g/L}$ ( $p < 0.001$ , one way ANOVA and Tukey test). Similarly, the concentration of 3,5,6-trichloro-2-pyridinol (TCPY) was significantly decreased during phase 2 compared with phase 1 or phase 3, $1.7 \pm 2.7 \mu\text{g/L}$ versus $7.2 \pm 5.8$ and $5.8 \pm 5.4 \mu\text{g/L}$ ( $p < 0.001$ , one way ANOVA and Tukey test). TCPY is a metabolite of chlorpyrifos.
<b>Conclusion</b>	Organic diets protect the children from organophosphorus pesticide exposure.
<b>Confounders adjusted for</b>	No
<b>Relevance for our assessment purpose</b>	Yes

<b>Reference</b>	<b>Curl, C. L., Fenske, R. A., &amp; Elgethun, K. (2003). Organophosphorus pesticide exposure of urban and suburban preschool children with organic and conventional diets. <i>Environmental Health Perspectives</i>, 111(3), 377-382</b>
<b>Study design and type</b>	Human, prospective observational study. Data collection from food diaries for 3 days, collection of urine for 24 hours.
<b>Objective</b>	To measure the concentration of organophosphorus pesticide metabolites in urine from children consuming mostly organic or mostly conventional diets.
<b>Number of participants, country and age</b>	21 and 18, USA, age 46 to 47 months.
<b>Baseline characteristics of study subjects/cells</b>	Healthy children (56-57% males).
<b>Exposure, substances, food (type and amount)</b>	Organophosphorus pesticide exposure through diets.
<b>Exposure; nature of the food (type and amount)</b>	Organic or conventional foods.
<b>Health outcome, a definition of the health outcome and how it was measured</b>	Exposure to pesticides through diet may be a health hazard.
<b>Follow-up period, drop-outs</b>	Urine collection for 24 hours.
<b>Statistical analysis</b>	Wilcoxon matched paired signed rank test, Mann-Whitney U-test.
<b>Results</b>	Dimethylthiophosphate (DMTP) was the major organophosphorus pesticide metabolite detected in almost 87% of all urine samples. The children consuming organic diets had lower concentration of total dimethyl metabolites in the urine than in the children consuming conventional diets (0.03 versus 0.17 $\mu\text{mol/L}$ , $p=0.0003$ , Mann Whitney U-test).
<b>Conclusion</b>	Consumption of organic foods may protect children from organophosphorus pesticide exposure.
<b>Confounders adjusted for</b>	No
<b>Relevance for our assessment purpose</b>	Yes

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