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RISK ASSESSMENT FOR INFANTS EXPOSED TO FURAN FROM READY-TO-EAT THERMALLY PROCESSED FOOD PRODUCTS IN POLAND

OCENA RYZYKA DLA NIEMOWLĄT NARAŻONYCH NA FURAN OBECNY W UTRWALANYCH TERMICZNIE DANIACH GOTOWYCH DO SPOŻYCIA W POLSCE

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ABSTRACT

Background. Thermal processes and long storage of food lead to reactions between reducing sugars and amino acids, or with ascorbic acid, carbohydrates or polyunsaturated fatty acids. As a result of these reactions, new compounds are created. One of these compounds having an adverse effect on human health is furan.

Objective. The aim of this paper was to estimate the infants exposure to furan found in thermally processed jarred food products, as well as characterizing the risk by comparing the exposure to the reference dose (RfD) and calculating margins of exposure.

Materials and methods. The material consisted of 301 samples of thermally processed food for infants taken from the Polish market in years 2008 – 2010. The samples included vegetable-meat, vegetables and fruit jarred meals for infants and young children in which the furan levels were analyzed by GC/MS technique. The exposure to furan has been assessed for the 3, 4, 6, 9 ,12 months old infants using different consumption scenarios.

Results. The levels of furan ranged from <1 µg/kg (LOQ) to 166.9 µg/kg. The average furan concentration in all samples was 40.2 µg/kg. The estimated exposures, calculated with different nutrition scenarios, were in the range from 0.03 to 3.56 µg/kg bw/day and exceeded in some cases RfD set at level of 1 µg/kg bw/day. Margins of exposure (MOE) achieved values even below 300 for scenarios assuming higher consumption of vegetable and vegetable-meat products.

Conclusions. The magnitude of exposure to furan present in ready-to-eat meals among Polish infants is similar to data reported previously in other European countries but slightly higher than indicated in the recent EFSA report. As for some cases the estimated intake exceeds the RfD, and MOE) values are much lower than 10000 indicating a potential health concern, it is necessary to continue monitoring of furan in jarred food and estimate of its intake by infants.

STRESZCZENIE

Wprowadzenie. Procesy cieplne, a także długie przechowywanie żywności powodują, że zachodzą w niej reakcje pomiędzy aminokwasami i cukrami redukującymi, oraz procesy z udziałem kwasu askorbinowego, węglowodanów i wielonienasyconych kwasów tłuszczowych. W wyniku tych reakcji powstają nowe związki chemiczne. Jednym z tych związków, wykazujących niekorzystne działanie na zdrowie człowieka, jest furan.

Cel badań. Celem pracy było oszacowanie wielkości narażenia niemowląt na furan występujący w utrwalanych termicznie przetworach, a także scharakteryzowanie ryzyka przez porównanie narażenia z wartością dawki referencyjnej (RfD) oraz obliczenie marginesów narażenia.

Materiał i metody. Materiał do badań stanowiło 301 próbek przetworów dla niemowląt pobranych w latach 2008 – 2010 z obrotu handlowego na terenie całego kraju. Wśród próbek znajdowały się dania warzywno-mięsne, warzywne oraz owocowe przeznaczone dla niemowląt i małych dzieci, w których zawartość furanu oznaczano metodą GC/MS. Narażenie na furan szacowano dla niemowląt w wieku 3, 4, 6, 9 i 12 miesięcy wykorzystując różne scenariusze żywienia.

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Wyniki. Oznaczona zawartość furanu mieściła się w zakresie od <1 µg/kg (LOQ) do 166,9 µg/kg. Średnia zawartość furanu we wszystkich próbkach wynosiła 40,2 µg/kg. Oszacowana wielkość narażenia, przy założonych różnych scenariuszach żywienia, mieściła się w granicach 0,03 – 3,56 µg/kg m.c./dzień i przekraczała dla niektórych przypadków wartość RfD. Marginesy narażenia (MOE) wyniosły nawet poniżej 300 dla scenariuszy zakładających większe spożycie produktów warzywnych i warzywno-mięsnych.

Wnioski. Oszacowana wielkość narażenia niemowląt w Polsce na furan pobierany z daniami gotowymi do spożycia jest zbliżona do wcześniejszych danych pochodzących z innych państw europejskich, lecz nieco wyższa od wyników najnowszego raportu EFSA. Biorąc pod uwagę, że w niektórych przypadkach oszacowane narażenie przekraczało wartość RfD (1 µg/kg m.c./dzień), a także fakt, że wartości MOE były znacznie mniejsze od 10000 wskazując na istnienie potencjalnego zagrożenia dla zdrowia, konieczne jest kontynuowanie badań monitorowych zawartości furanu w żywności dla niemowląt i szacowanie narażenia na ten związek.

INTRODUCTION

Furan (C_4H_4O) (CAS No. 110-00-9) is a low molecular weight and highly volatile compound consisting of a five-membered aromatic ring with one oxygen atom with boiling point of 31.36°C, which implies its behavior in different media, including food. In industry furan is used as an intermediate product in the synthesis of numerous polymers and copolymers [5]. Its presence was confirmed in heat-treated food.

For the first time furan was identified in volatile components of coffee at the end of thirties of the twentieth century [20], which was later confirmed by subsequent studies. Further researches, in 1979 led to the publication of the report from the studies concerning the occurrence and formation of furan and its derivatives in food [23]. Previously, the only considered way by which furan was formed in food was *Maillard* reaction with participation of aminoacids and reducing sugars (nonenzymatic browning sugars). However, on the basis of recent studies it was proved that it can also be formed by thermal decomposition of carbohydrates, thermal oxidation and thermal degradation of polyunsaturated fatty acids (PUFA), ascorbic acid and derivatives thereof. The compounds that probably are also responsible for the formation of furan in food include triglycerides, organic acids and carotenes [4, 13-15, 21, 22, 26, 28, 36].

Genotoxic mechanism of furan was confirmed in numerous tests which allowed to detect different types of mutations [25, 30], caused by *cis*-2-butene-1,4-dial as a essential cytotoxic reactive metabolite of furan [3], which can *in vitro* react with deoxyribonucleotides to form unstable DNA adducts [1]. According to the European Food Safety Authority (EFSA), the available data about mechanism of action of furan indicate its carcinogenic properities due to its genotoxic activity [5]. The UK Committees on: Toxicity Mutagenicity Carcinogenicity of Chemicals in Food, Consumer Products and the Environment in 2005 concluded that furan should be regarded as an *in vitro* mutagen, but that there was insufficient evidence to reach a conclusion on the available *in vivo* mutagenicity data [31].

Furan is carcinogenic to rats and mice, showing a dose-dependent increase in hepatocellular adenomas and carcinomas in both sexes. In rats of both sexes, there was also a dose-dependent increase in the incidence of mononuclear leukemia, and a high incidence of cholangiocarcinoma of the liver, even at extremely low doses [16]. Chronic exposure to furan can result in hepatocytes proliferation which can lead to liver tumors [2, 5]. In 1995, furan has been classified by the International Agency for Research on Cancer (IARC) to the group 2B as a possibly carcinogen for humans [17].

Furan has low polarity, can pass through biological membranes, and is rapidly absorbed and extensively metabolised after ingestion by rats and mice. It can get into human body by ingestion and inhalation while preparing food [16].

Report of the Joint FAO/WHO Expert Committee on Food Additives (JECFA) in 2010 provides the benchmark dose lower limit $BDML_{10}$ equal to 0.96 mg/kg bw/day which made possible to set a margin of exposure (MOE) in humans (expressed as the ratio between the estimated $BMDL_{10}$ and estimated exposure). The $BDML_{10}$ value was derived from long-term carcinogenicity studies on mice where hepatocellular adenomas and carcinomas were chosen as tumors associated with administration of furan. Comparison of dietary exposures of 0.001 mg/kg bw per day, to represent the average dietary exposure to furan for the general population, and 0.002 mg/kg bw per day, to represent the dietary exposure to furan for consumers with high dietary exposure covering also dietary exposure for children, with above $BMDL_{10}$ gives MOEs of 960 and 480 depending on the exposure scenario. The Committee considered that these MOE's indicate a human health concern for a carcinogenic compound that might act *via* a DNA-reactive genotoxic metabolite [18].

It should be emphasized however, that the risk of carcinogenic effects in infants and young children may be higher than in adults, because the rate of cell proliferation in the developing liver may increase the subsequent risk of cancer [12].

FDA has estimated the exposure to furan for the general population (above 2 years old) [34] on average level of 0.26 µg/kg bw/day, and for 90 percentile at 0.61 µg/kg bw/day. The average exposure of infants and children up to one year to furan found in food according to FDA was 0.41 µg/kg bw/day, and for the 90th percentile – 0.99 µg/kg bw/day. Exposure resulting from the consumption of convenience food for infants and young children (infant formula), as the only source of food was estimated as high as 0.9 µg/kg bw/day.

Many pathways of furan's formation led to its presence in food as a result of heat treatment in closed vessel (for example during pasteurization), as well as in open vessels (cooking, frying or baking). The presence of furan has been confirmed in more than 300 products. Despite the high volatility of furan and due to a low boiling point which both create serious analytical difficulties, it is possible to quantify even trace concentrations in food samples. The highest content (except coffee), up to 200 µg/kg, were found in baked products, such as cookies, chips and crispy bread. Content of furan was correlated with an increase in browning of the final product. FDA published on their website the results of the concentrations of furan in various food presented between 2004 and 2006 [33]. They were consistent with the data published by the Swiss Federal Office of Public Health in 2004 and 2007 [27, 38].

Definitely, the highest content of furan was found in coffee, where it reached the value up to 6000 µg/kg [38]. That high concentrations were found in ground coffee, much lower content were in instant coffee (2150 µg/kg) and coffee ready to use called as mix 2 in 1, 3 in 1 (100 µg/kg). Furan can also be found in the indoor air in kitchens [8] and the data show that the highest amount of furan can be inhaled during addition of hot water to coffee in a cafetiere, during pan-frying and baking some food in the oven.

In 2009 and 2011 EFSA [7, 9] published the results from the EU countries concerning the content of furan in various food products, including ready-to-eat products for infants and young children. Furan levels in most of the products were comparable with the previous results. The EFSA report from 2004 include the results of 273 children's meals (vegetable, vegetable-meat, and fruit), showing that furan may reach levels up to 112 µg/kg [5]. On the basis of recently published studies it can be assumed that among the ready-to-eat jarred meals for children, the highest furan levels were found in vegetable and vegetable-meat meals and the lowest in fruit based meals. Other authors in their studies [7, 19] presented similar data on food consumption, for example EFSA [5] reports that a consumption of 234 g of jarred food results in furan exposure ranging from 0.2 to 26.2 µg furan per day.

While assessing exposure to furan it should be taken under consideration that due to the low body weight and not fully formed detoxification mechanism, infants and young children are particularly vulnerable to the potential harmful effects of this compound.

The aim of this study was to assess the exposure of infants to furan found in ready-to-eat jarred food products, which constitute a dominant part of their diet as well as characterizing the risk by comparing the exposure to the reference dose (RfD) and calculating margins of exposure.

MATERIAL AND METHODS

The material consisted of 301 samples of thermally processed jarred food products for infants taken from the Polish market as part of monitoring studies from 2008 to 2010. Products originated from four major manufacturers and differed by brand name, composition, and age recommendation. The samples included vegetable-meat, vegetables and fruit jarred meals for infants and young children aged between 4 and 18 months. The results of furan analysis in these products were published earlier [24, 29]. From among numerous methods, the gas chromatography coupled with mass spectrometry (GC/MS) with a head-space technique (HS) has been chosen as a method based on the published by the FDA [32]. This method has been validated and was accredited by the Polish Centre for Accreditation (PCA), Certificate No AB 509. Selected validation parameters were as follows: the limit of detection (LOD) 0.5 µg/kg, the limit of quantification (LOQ) 1.0 µg/kg, recovery 70-120% and expanded measurement uncertainty (k = 2) – 19.7%. As internal standards deuterated furan has been used so the recovery was determined for each sample due to the low boiling point and related difficulties in analysis.

The exposure to furan has been assessed for the 3, 4, 6, 9 ,12 months old infants using different consumption scenarios.

RESULTS

Analytical studies

The presence of furan was detected in 300 of 301 analyzed samples and its levels ranged up to 166.9 µg/kg. Among all the products, the fruit based meals revealed a significantly lower content of furan than vegetable and vegetable-meat products. The average furan concentration for all samples was 40.2 µg/kg with median equal to 41.1 µg/kg, while the average recovery was 92.5%. The Table 1 summarizes some basic statistical data characterizing the furan analysis in various groups of products.

The child age is of essential importance in the exposure evaluation, since it is related to the magnitude of consumption based on the weight and type of the product. The Table 2 summarizes the statistical data characterizing the results for all products, grouped by recommended age.

Table 3 shows the average concentration of furan in analyzed products with regard to age recommendation and depending on the jarred food category. This approach should facilitate to indicate the different types of scenario in terms of exposure to furan.

Risk assessment

The exposure to furan present in ready-to-eat jarred products for infants has been assessed for the 3, 4, 6, 9 ,12-months old infants. To qualify the product to the particular age group the manufacturer's recommendations, shown on the label, were used and followed in the first columns of Tables 2 and 3. The age relevant body weights (50th percentile) of boys and girls were taken from the growth charts (Table 4) and used for infants exposure assessment to diet derived furan.

For exposure assessment purposes, three scenarios (Table 5) based on infants nutrition schemes displayed on the product label were taken into account. Each of the scenarios assumed that the child eats the ready-

Table 1. Furan levels in samples of ready-to-eat jarred meals for infants and small children
Zawartość furanu w próbkach dań gotowych do spożycia dla niemowląt i małych dzieci

Ready-to-eat food	Number of samples	Mean (μg/kg)	Recovery (%)	Minimum (μg/kg)	Maximum (μg/kg)	Median (μg/kg)
Vegetable-meat	208	44.2	92.6	5.1	115.6	42.9
Vegetable	45	60.1	91.3	24.0	166.9	54.0
Fruit	48	4.0	93.5	< 1.0	17.4	3.6

Table 2. Furan levels in samples of ready-to-eat jarred meals grouped by recommended infants age recommendation
Zawartość furanu w daniach gotowych do spożycia wg kategorii wiekowej

Age (month)	Number of samples	Mean (μg/kg)	Recovery (%)	Minimum (μg/kg)	Maximum (μg/kg)	Median (μg/kg)
> 4	59	36.8	92.3	< 1.0	166.9	37.9
> 5	72	41.4	93.9	3.8	71.1	40.9
> 6	65	42.4	93.9	1.7	91.4	42.0
> 7	5	6.5	95.4	5.1	10.3	5.7
> 8	44	45.2	89.2	11.2	76.9	44.1
> 9	25	31.3	93.1	1.8	141.5	13.2
> 11	1	8.1	85.7	8.1	8.1	8.1
> 12	29	46.6	89.9	13.1	115.6	45.4
> 18	1	41.1	108.6	41.1	41.1	41.1
all samples	301	40.2	92.5	< 1.0	166.9	41.0

Table 3. Furan levels in ready-to-eat jarred meals depending on age recommendation and food category
Zawartość furanu w daniach gotowych do spożycia w zależności od kategorii wiekowej i rodzaju produktu

Age (month)	Number of samples	Mean (μg/kg)	Recovery (%)	Minimum (μg/kg)	Maximum (μg/kg)	Median (μg/kg)
Vegetable-meat food						
over 5	70	42.5	93.8	13.6	71.1	41.3
over 6	50	49.1	94.4	14.9	91.4	46.4
over 7	4	6.9	96.4	5.1	10.3	6.0
over 8	43	44.7	89.6	11.2	76.9	43.8
over 9	9	41.0	93.5	13.2	84.6	38.8
over 11 (11-18)	31	45.2	90.3	8.1	115.6	45.4
Vegetable food						
over 4	36	58.3	91.3	24.0	166.9	56.5
over 6	6	43.7	89.2	36.5	57.6	41.5
over 9	3	114.3	94.8	82.0	141.5	119.3
Fruit food						
over 4	23	3.1	93.8	< 1.0	6.7	2.4
over 5	2	3.9	96.1	3.8	4.0	3.9
over 6	9	4.2	94.0	1.7	9.2	3.5
over 7 (7-9)	14	5.4	92.3	1.8	17.4	4.0

Table 4. Data on body weight of boys and girls according to age
Dane dotyczące masy ciała chłopców i dziewczynek w zależności od wieku

Age (month)	Boys (kg)	Girls (kg)
3	6.5	5.8
4	7.2	6.5
6	8.3	7.5
9	9.4	8.6
12	10.2	9.4

-to-eat jarred products in addition to mother's milk or milk formula.

The best for infants nutrition model of minimum exposure assumes that the child is fed as long as possible with mother's milk or milk formula. This situation is possible in the case of 3 and 4-month-old infants. Ready-to-eat jarred food are introduced into the diet later in the following months. Other assumptions take into account the recommendations of the Panel of Experts appointed by the National Consultant in Paediatrics [37], as well as the recommendations by the producer of ready-to-eat jarred products, as they are applied in practice. They assume extension of diet after 4-5 months with 3-4 teaspoons of vegetables pulp. Afterwards the fruit meals are introduced into diet, first comprising of one component and later are expanded into more diverse products. After 6 months, a menu should include meals containing vegetables and meat, and child is expected to eat fruits between meals. After 7 months larger quantities of food and more diversified food including heterogeneous meals are suggested, although continuation of foregoing feeding patterns becomes also frequent practice. After 8 months new components are introduced both vegetable, vegetable-meat, and fruit meals. With the development of the child, larger portions should be given. In this study, example scenarios take into account general recommendations on infants nutrition and different needs of particular infants. Estimated exposure to furan and MOE expressed as the ratio between the $BMDL_{10}$ applied recently by JECFA and estimated exposures based on three suggested scenarios are presented in Table 5.

Table 5. The exposure of infants to furan from ready-to-eat jarred meals based on various nutrition scenarios
Narażenie niemowląt na furan pobierany z daniami gotowymi do spożycia na podstawie różnych scenariuszy żywienia

Age of infants (month)	Assumed model of nutrition	Average weight of the jar (g)	Furan intake (µg/day)	Exposure to furan – boys / girls (µg/kg bw/day)	MOE boys / girls
Scenario I					
3	only mother milk or infant formula	—	—	— / —	— / —
4	only mother milk or infant formula	—	—	— / —	— / —
6	1/2 vegetable food + 1/2 fruit food	166.7	3.99	0.48 / 0.53	2000 / 1811
9	1 vegetable food + 1/2 fruit food	175.0	20.47	2.17 / 2.38	442 / 403
12	1/2 vegetable-meat food + 1 fruit food	242.5	6.79	0.67 / 0.72	1433 / 1333
Scenario II					
3	only mother milk or infant formula	—	—	— / —	—
4	1/2 fruit food	127.4	0.20	0.03 / 0.03	32000 / 32000
6	1/2 vegetable food	166.7	3.64	0.44 / 0.49	2182 / 1959
9	1 vegetable food + 1 fruit food	175.0	20.95	2.23 / 2.44	430 / 393
12	1 vegetable food + 1/2 vegetable-meat food + 1 fruit food	242.5	33.45	3.28 / 3.56	293 / 270
Scenario III					
3	1/2 fruit food	127.4	0.20	0.03 / 0.03	32000 / 32000
4	1/2 vegetable food + 1/2 fruit food	127.4	3.91	0.54 / 0.60	1778 / 1600
6	1 vegetable food + 1 fruit food	166.7	7.98	0.96 / 1.06	1000 / 906
9	1 vegetable-meat food + 1 fruit food	175.0	8.12	0.86 / 0.94	1116 / 1021
12	2 vegetable-meat food + 1 fruit food	242.5	23.23	2.28 / 2.47	421 / 389

DISCUSSION

The assessment of the exposures to furan based on the scenarios showed in the Table 5 apply the deterministic approach that made possible to estimate the health risk in the most frequently occurring situations. The health risk in this case was related to the estimated value reflecting how much the reference dose (RfD) for furan (1 μg/kg bw/day) was exceeded when the particular scenarios were applied. The RfD has been set by US EPA for non-carcinogenic effects in 1989 [11]. The US reference doses are often compared to FAO/WHO's ADI values.

The variables in this case were the constituents of recommended meals for the age groups and the amounts consumed. This approach allowed to reveal that infants which were the least exposed to furan belonged to 3 to 4-month-old age group and additionally to 12-month-old infants in scenario I, and 9-month-old in scenario III. The most exposed to furan were 9-month-old infants fed according to scenario I, and 12-month-old infants in scenario II and 12-month-old infants in scenario III.

The exposure estimated for infants and young children on the basis on own studies ranged between 0.03 – 3.56 μg/kg bw/day with frequently occurred maximum exposure above the RfD set for furan. It should be emphasized that these values are almost identical to those reported by EFSA in 2004 for 6-months old infants (0.03 – 3.5 μg/kg bw/day) [5]. However, the latest EFSA Report of 2011 [9] shows that infants exposure to furan from all food sources decreased to 0.09 – 0.22 μg/kg bw/day with a high exposure estimate at the 95th percentile of 0.97 μg/kg bw/day. The Report point out that these values were estimated based on the limited food consumption information for infants (only two surveys). Similarly, recently published Report of Norwegian Scientific Committee for Food Safety indicates lower exposure of 6- and 12-months old infants – consumers of jarred baby foods – equal to 0.18 and 0.56 μg/kg bw/day (means), and 0.45 and 1.30 μg/kg bw/day (95th percentiles), respectively [35].

Higher exposure values indicating that the RfD could be exceeded may also be due to the methodology of calculation and certainly be true for some scenarios. On the contrary, if one assumes an average content of furan in all tested samples, without grouping by purpose or type of processed product, the estimated exposure would only slightly exceeds the RfD reaching a maximum of 3.56 μg/kg bw/day for scenario II for 12-month-old girls. This proves that more refined approach to risk assessment should be applied in order to indicate groups of children at the highest risk expressed for example as a percent above the toxicological reference value.

In 2005 the Scientific Committee has been asked by the EFSA to propose a harmonized approach for the risk assessment of substances that have both genotoxic and carcinogenic properties [6, 10]. In light of recent toxicological data furan undoubtedly belongs to such substances. The Committee recommended using an approach known as the margin of exposure. As the RfD value is based on 30 years old studies with non-carcinogenic effect as a end-point chosen to derive NOAEL, it seems to be more appropriate to use MOE in order to characterize the risk connected wit furan exposure. The MOE values calculated in this study and presented in table 5 are consistent with value of 480 calculated by JECFA. Independently of age of the infant, consumption of one jar of vegetable meal results in MOE below 1000 with the lowest value (270) for 12-months old girls. The MOE values calculated in Norwegian Report are even lower – for 6-months old infants the values are equal to 111 and 44 for mean exposure, and high exposure, respectively, and for 12-months old infants are 35 and 15 for mean exposure, and high exposure, respectively. However it is worthy to note, that another $BMDL_{10}$ value has been derived and used for MOE calculations [35].

It is accepted that in general, margin of exposure of 10000 or higher, if it is based on the $BMDL_{10}$ from an animal study, and taking into account overall uncertainties in the interpretation, would be of low concern from a public health point of view. Therefore, results obtained in this study indicate a potential health concern and may be considered as high priority for risk management actions.

CONCLUSIONS

1. The children exposure in Poland to furan in ready-to-eat jarred meals does not differ from reported previously from other European countries and is almost identical to the data published by EFSA in 2004, but higher to recent EFSA Report of 2011.
2. The methodology proposed in this study allows for independent evalulation of the exposure taking into account different scenarios.
3. Since in this work we revealed the possibility that some groups of infants and small children may be exposed to furan above the toxicological reference value, it is postulated that the continuation of the monitoring of infants and young children exposure to furan from processed food products is necessary to generate more data for more refined risk assessment.
4. The MOE values less than 1000 obtained in this study indicate a potential health concern and may be considered as high priority for risk management actions. Research on technological processes allowing reduction of furan in commercial baby foods should be conducted.

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