AGENCE FRANÇAISE
DE SÉCURITÉ SANITAIRE
DES ALIMENTS

Maisons-Alfort, 29 January 2010

OPINION

of the French Food Safety Agency on the critical analysis of the results of a developmental neurotoxicity study of bisphenol A together with other recently-published data on its toxic effects

1. REVIEW OF THE REQUEST

On 20 October 2009 the French Food Safety Agency (AFSSA) issued an internal request to analyse the results of a developmental neurotoxicity study of bisphenol A in rats, together with other recently-published data on its toxic effects.

2. CONTEXT AND QUESTIONS RAISED

AFSSA requested on 10 September 2009 and received on 8 October 2009 the full report of a study commissioned by the American Chemistry Council addressing health concerns raised by Northern European countries in the context of the European programme for assessing existing chemical substances (under regulations that preceded REACH[1]), by the NTP[2] and by the Canadian government. The study was performed in compliance with Test Guideline 426 issued by the OECD[3] which is designed to detect neurological, morphological and behavioural anomalies (learning or memory difficulties, etc.) from birth to adulthood, caused by maternal exposure (during gestation and lactation) to bisphenol A.

In addition, several studies in the literature suggest that effects occur at doses lower than the No Observed Adverse Effect Level (NOAEL) set by EFSA[4] (5 mg/kg b.w./day) for calculating a Tolerable Daily Intake (TDI) of 0.05 mg/kg b.w./day.

In the European and international context, AFSSA decided to analyse the results of these studies (including about fifty articles and scientific reports published in 2008 and 2009) and issued an internal request on 20 October 2009 to respond to the following questions:

1) Does the developmental neurotoxicity study of bisphenol A, performed in compliance with OECD Test Guideline 426, show effects following the exposure of litters during gestation and suckling?
2) Does the study disprove the toxicity of this compound at low doses on neurological and behavioural development? Do recent findings in the literature confirm the occurrence of effects suggesting a risk to public health at very low doses? Should these findings lead to a modification of the NOAEL used for calculating the TDI?
3) More generally, is the methodology for risk assessment, based on the notion of a TDI, the most suitable for application to endocrine disruptor compounds, such as bisphenol A?

[1] REACH: European Regulation (EC) 1907/2006 on the Registration, Evaluation, Authorisation and restriction of CHemicals (in force since 1 June 2007).
[2] NTP (National Toxicology Program). NTP-CERHR Monograph on the Potential Human Reproductive and Developmental Effects of Bisphenol A, September 2008, NIH Publication No. 08 – 5994, 321p.
[3] OECD: Organisation for Economic Cooperation and Development
[4] European Food Safety Authority

3. EXPERT ASSESSMENT METHOD

The 'Bisphenol A' Working Group (WG), set up on the initiative of the Director General of the French Food Safety Agency, was appointed to conduct this assessment.
The 'Bisphenol A' WG was made up of experts from the Scientific Panels on 'Chemical and physical contaminants and residues', 'Food contact materials', 'Plant protection products: chemical substances and preparations' and 'Fertilizers and growing media' and an outside expert from INRA-Toulouse.

The "Réseau Environnement Santé" was invited to present a summary of the literature on this topic on 01 December 2009.

On the basis of the WG's report and after consulting the Scientific Panels on 'Chemical and physical contaminants and residues ' (CES RCCP) and 'Food contact materials' (CES MCDA), which met on 13 and 21 January 2010, AFSSA is issuing the following conclusions and recommendations.

4. DISCUSSION

QUESTION 1

The toxicity study in rats, performed in compliance with OECD Test Guideline 426, covering a wide range of doses of bisphenol A (0.15, 1.5, 75, 750 and 2250 ppm per day in the mother's feed, from the first day of gestation to the last day of suckling on postnatal day 21), revealed no adverse effects on the development of the nervous system at the No Observed Adverse Effect Level (NOAEL) for the mother (less than or equal to 75 ppm). The only sign of toxicity observed in the mother was a reduction in weight gain of 9.5 and 22.4% respectively at doses of 750 and 2250 ppm, solely during gestation, with a concomitant reduction in feed consumption. At these doses, convulsions were observed in some of the pups at the age of 11 days. The historic data show a lower incidence of cases of convulsion, and only in females. Nevertheless, since no convulsions occurred in the course of a complementary test (carried out only with the highest dose and for 11 days), the authors did not take the convulsions into consideration and proposed a NOAEL corresponding to the highest dose tested (2250 ppm).

The experts consider that this study does not show any neurotoxic effect on the litters at the no effect doses for the mother but that it does not allow a definitive conclusion for the highest doses, bearing in mind the absence of any investigation on the origin of the convulsions observed in some rats.

QUESTION 2

The study in rats, performed in compliance with OECD Test Guideline 426, does not reach a formal conclusion that there are no effects at doses lower than the NOAEL of 5 mg/kg b.w./day. Indeed, in the absence of any measurement of plasma levels, it is not possible to determine the internal dose of exposure to bisphenol A of rats *in utero* and during suckling.
Furthermore, the exposure of the animals to other endocrine disruptors was not sufficiently controlled under the experimental conditions (for example, the presence of phytoestrogens in their diet, or of bisphenol A or phtalates in plastic cages) such that interference with the possible effects of bisphenol A at very low doses cannot be excluded.
AFSSA is ready to examine any complementary information received that may shed light on the issues raised by the answers to Questions 1 and 2.

In the studies of the literature, the effects observed at very low doses concern subtle modifications of functions (neurological, motor or sensorial functions), hormone balance or metabolism and should be interpreted as warning signals because it has not been established whether or not they have harmful consequences for human health.
However, these studies suffer from considerable methodological bias and cannot be used to establish any dose-response relationship nor to determine a NOAEL on which to base a TDI.

In the current state of knowledge, it is not possible to correlate the human biomonitoring data with the effects observed *in vivo* in animals in laboratory conditions, because of insufficient toxicokinetic data.

QUESTION 3

The TDI is the maximum quantity of a contaminant that can be consumed daily over an entire lifetime without the risk of harmful effects on human health.
In the case of compounds that are endocrine disruptors, which can have different effects depending on the development stage (critical exposure windows during which harmful effects can appear, in particular the perinatal period), the TDI does not seem to be the most suitable approach for risk assessment.

Furthermore, the OECD Test Guideline 426 does not seem entirely suitable for characterising subtle effects on the nervous system, such as may be observed with endocrine disruptors and bisphenol A in particular.

5. RECOMMENDATIONS

It is necessary to determine the significance in terms of health safety of the warning signals observed in the *in vitro* and *in vivo* studies at doses lower than the NOAEL of 5 mg/kg b.w./day. In the meantime, and taking into account the fact that the significance of these signals for human health is uncertain, the relevance of increasing the safety factor of the TDI should be debated and the other sources of exposure to bisphenol A than food contact materials should be investigated..

Based on the particular case of bisphenol A, the experts are issuing the following recommendations for toxicity studies and the assessment of the health risk related to endocrine disruptors:

- Concerning the toxicity studies designed to establish toxicological reference values (in particular the guidelines for regulatory toxicology tests), these should include:
    - toxicokinetic parameters and particularly plasma and/or urine concentrations;
    - hormonal analysis (concentrations of hormones and their metabolite(s) in the blood and urine);
    - a study of the effects on physiological functions identified as critical, depending on the development stage at the time of exposure;
    - consideration of methodological bias, such as the effects of diet (the presence of phytoestrogens in soy-based products), polycarbonate cages, the composition of the drinking water given to the animals, the bedding (which may contain mycotoxins, terpenes, polyphenols etc.).
- The experts stress that it is indispensable to test several doses in order to determine a dose-response relationship.
- A methodology should be developed for assessing the potential health risks of very low doses of endocrine disruptors.
  In the meantime, the risk assessment approach may be based on the calculation of margins of exposure[5] (MOE), which takes into account the particular sensitivity of humans at certain stages of life and avoids the need to determine a safety factor *a priori*. However, it is difficult to apply this approach to bisphenol A before the significance of the warning signals has been established.

6. AFSSA'S CONCLUSION

Bisphenol A has been used for many years in food and water contact materials.

The assessment of dietary exposure fell outside the scope of this request, but the data analysed enable AFSSA to propose an estimate for the exposure of infants.

[5] The MOE can be calculated by comparing data from the studies on laboratory animals with human data, for different population groups (pregnant women, infants, adults, men/women) on the basis of either dietary intake or biomonitoring data.

On the basis of daily consumption of milk[6] of 174 ml/kg of body weight (b.w.), the data (from the USA, Japan and Canada) show that infants would be exposed to:

- 0.33 – 1.27 µg of BPA/kg bw/day from breast milk[7] (for mean and maximum concentrations of total bisphenol A);
- 0.20 – 2.1 µg of BPA/kg bw/day from infant formula milk[8] (through migration from the packaging);
- 0.017 – 0.12 µg of BPA/kg bw/day through migration from the baby bottle, under realistic conditions of use[9].

Toxicity studies performed in compliance with international standards have not so far demonstrated any risk to health at current levels of exposure. Regardless of the feeding method, exposure of infants is well below the TDI based on these studies.

However, recent publications, whose methodology does not authorise any formal conclusions, mention warning signals following *in utero* and postnatal exposure at doses lower than that on which the TDI is based.
The consequences for human health of these warning signals have not been clearly determined.
Furthermore, if these warning signals concern the oestrogenic activity of bisphenol A, it is clearly essential to understand the mechanisms behind the effect of bisphenol A on humans, who are also exposed to other compounds with an oestrogenic activity, whether of chemical or natural origin, found in certain foods.

In this context, AFSSA will continue its expert assessment work, jointly with the international network of health agencies, to ascertain the significance for human health of these warning signals and thus be able to propose new methodologies for assessing the risks related to very low levels of bisphenol A and more generally of endocrine disruptors.
AFSSA recommends collecting data in France on the presence of bisphenol A in breast milk, in infants and in infant formula.
The Agency also recommends investigating sources of exposure to bisphenol A other than food contact materials.

These new data will be important for informing consumers and for helping the authorities to take measures appropriate to the risk.

The Director General

Marc MORTUREUX

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[9] Ehlert *et al.* (2008), De Coensel (2009), Kubwabo *et al.* (2009)

KEYWORDS

Bisphenol A, toxicity, warning signals, nervous system, development, endocrine disruptor, exposure window

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