This is provisional English translation of an excerpt from the original full report.

Risk Assessment Report

Foods highly containing diacylglycerol

(Novel Foods and Food Additives)

Food Safety Commission of Japan (FSCJ)

March 2015

ABSTRACT

The Food Safety Commission of Japan (FSCJ) conducted a risk assessment on foods highly containing diacylglycerol (DAG) (hereinafter referred to as DAG foods), based on results from various studies.

The safety of DAG foods, as foods for specified health use, was once evaluated by the Pharmaceutical Affairs and Food Sanitation Council of the Ministry of Health, Labour and Welfare (MHLW). On September 11, 2003, FSCJ approved the evaluation of the MHLW's council to be appropriate.

Subsequent MHLW's two-stage carcinogenicity studies on edible oils highly containing DAG (hereinafter referred to as DAG oils) conducted from September 2005 to February 2009 supported the MHLW's judgment "No concern relevant to human health even though all the edible conventional oils were substituted with DAG oils". FSCJ continued to conduct also a risk assessment of DAG oils focusing on the minor constituent, glycidol fatty acid esters (GE).

DAG oils and related products have been, however, retracted from the market in September 2009 in Japan. DAG foods are thus unlikely to expose people, and no further data necessary for the assessment are available on the consumption of DAG foods (period, amount, age, etc.) and varieties of confounding factors in lifestyle. Lack of individual data on the exposure makes it difficult from the reliable estimation of lifetime cancer risk. The food safety assessment thus could not be completed.

Current scientific considerations on the possible adverse effects of DAG oils and GE as contaminants in food oils are summarized in the appendices.

Appendix 1: On DAG oils

- DAG oils showed no tumor promoting activity after the oral administration in the oral cavity and tongue, esophagus, forestomach and colon of mice.
- DAG oils showed no carcinogenicity in the mammary gland in animal studies.
- Both the experiments described above were done at much higher doses compared with human daily consumption.

- As the results, the tumor promoting activity of DAG oils, once concerned, was denied. Adverse effects of DAG oils were not confirmed in animal experiments, although DAG oils contain GE as contaminants.
- Hence, FSCJ judged that tumor promoting risk in human is negligible from the daily consumption of DAG oils in foods.

(Full version is available in Japanese at: http://www.fsc.go.jp/fsciis/attachedFile/download?retrievalId=kya20050920001&fileId=200.)

Appendix 2: On Glycidol and its fatty acid esters (GE)

- DAG oils were suspected to contain trace amounts of GE as contaminants. The genotoxic carcinogenicity was not denied for the metabolite of GE, glycidol.
- Studies, however, showed the rather weak genotoxicity of GE than glycidol. The incidence and multiplicity of tumors in carcinogenicity studies were rather lower with GE than glycidol.
- The level of GE in the currently available edible oils and food products is maintained as minute. For this exposure estimation, all of the GE were assumed to be converted to the equimoles of glycidol in the body. Even though the possible overestimation, a certain level of margin of exposure (MOE), slightly lower than 10,000, was allocated.
- These data suggest no apparent adverse effects due to the consumption of edible oils currently available. GE exposure levels, however, should be kept as low as possible according to the principle of ALARA (As Low As Reasonably Achievable).
- For future risk assessments, detailed data on GE such as the toxicokinetics and toxicity on individual GE substances, as well as human exposure, would be helpful.

(Full version is available in Japanese at: http://www.fsc.go.jp/fsciis/attachedFile/download?retrievalId=kya20050920001&fileId=200. The executive summary is available in the accompanying annex.)

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Annex to the Risk Assessment Report of "Foods highly containing diacylglycerol"

Considerations on glycidol and its fatty acid esters in foods

(Novel Foods and Food Additives)

Food Safety Commission of Japan (FSCJ)
March 2015

ABSTRACT

As a part of a safety assessment of foods highly containing diacylglycerol (DAG), the Food Safety Commission of Japan (FSCJ) conducted a safety assessment of glycidol and its fatty acid esters (GE) in foods based on results from various studies. The level of GE in the currently available edible oils is maintained as minute, and even if all of the GE were assumed to be converted to the equimoles of glycidol in the body, the extra risks of tumor incidence were estimated to be very low, and a certain level of margin of exposure (MOE), slightly lower than 10,000, was allocated. Additionally, so far no reports on adverse effects of consuming oils containing GE on human health have been reported. While these data suggest no apparent adverse effects due to the consumption of edible oils currently available, the genotoxic carcinogenicity of glycidol was not denied. Therefore, GE exposure levels should be kept as low as possible according to the principle of ALARA (As Low As Reasonably Achievable).

Introduction

As a part of a safety assessment of foods highly containing diacylglycerol (DAG), the Food Safety Commission of Japan (FSCJ) conducted a safety assessment of glycidol and its fatty acid esters (GE) in foods based on results from the following studies: pharmacokinetics, genotoxicity, acute toxicity, repeated dose toxicity, carcinogenicity, and reproductive and developmental toxicities.

1. Pharmacokinetics

Pharmacokinetics data are available only for glycidol linoleate among the GE. No apparent differences are expected in the rates of hydrolysis of glycidol linoleate with other long chain fatty acid esters (glycidol palmitate, stearate, oleate or linolenate). Experimental data indicate that glycidol linoleate is absorbed rapidly into the blood to appear mainly in the form of glycidol in rats. It may thus be reasonably assumed that GE are invariably converted to equivalent amount of glycidol in the body. Bioavailability of glycidol in *Macaca fascicularis* has been reported to be lower than in rats after the oral administration of GE, suggesting species differences in the bioavailability of GE assessed as glycidol. However, because of the unavailability of human pharmacokinetics data, FSCJ judged that it is appropriate to conduct the current assessments following the conservative procedure on the basis of data from studies in rats.

2. Toxicities

Considerable amounts of evidence are available on genotoxicity of glycidol, particularly on the glycidol-induced DNA damage and gene mutation. Carcinogenicity tests have shown increased incidences of tumors attributable to glycidol exposure in rats and mice. A study in hamsters also showed weak carcinogenicity of glycidol. These results suggest glycidol as a genotoxic carcinogen. In addition, some exposure-related effects were observed in the glycidol studies on repeated dose toxicity, reproductive and developmental toxicity, and immunotoxicity. Studies, however, showed the rather weak genotoxicity of GE than glycidol. In addition, although data are limited on the carcinogenicity of GE, results of carcinogenicity tests currently available are consistent with the idea that GE are rather weak carcinogens compared with glycidol. In fact, the incidence and multiplicity of tumors in carcinogenicity studies were rather lower with GE than glycidol.

Taking genotoxic carcinogenicity as the most discernible endpoint, FSCJ considered it appropriate to apply the worst-case scenario that GE are fully converted to glycidol during absorption process and are then available in the body.

3. TDI and Unit Risk

Since involvement of genotoxic carcinogenicity of glycidol could not be excluded, both tolerable daily intake (TDI) and Unit Risk estimations are applied for the assessment of glycidol.

No-observed-adverse-effect level (NOAEL) for carcinogenicity was not be specified in carcinogenicity tests in rats and mice by National Toxicological Program (NTP) (1990). Therefore Benchmark Dose (BMD) approach rather than NOAEL was used for the assessment, and Benchmark Dose Lower Confidence Limit 10% ($BMDL_{10}$) was thus obtained as 1.6 mg/kg bw/day. The TDI was also specified to be 1.6×10^{-3} mg/kg bw/day, applying an uncertainty factor of 1000 (100 as default and additional 10 for the severity (carcinogenicity)) to the $BMDL_{10}$.

The exposure levels corresponding to the extra risks of tumor incidence of 10^{-4}, 10^{-5} and 10^{-6}, calculated from the $BMDL_{10}$, were as 1.6×10^{-3}, 1.6×10^{-4} and 1.6×10^{-5} mg/kg bw/day, respectively, for Unit Risk estimation.

4. Exposure Assessment

The exposure assessments of GE were conducted on the assumption that all of the GE in the relevant edible oils were converted to the equimoles of glycidol in the body.

The daily intakes of GE are estimated in the following two populations, based on the results of National Health and Nutrition Survey. One is the total population. The other is a male population aged from 15 to 19, who showed the maximum ingestion of edible oils in average. As the results, the average and maximum daily intakes of glycidol derived from GE were estimated to be 9.0×10^{-5} and 1.5×10^{-4} mg/kg bw/day, respectively, and their Margins of Exposure (MOEs) of GE were calculated as approximately 17,800 and 10,900, respectively.

It is necessary to note that edible oils used in manufacturing of processed foods are not counted in the estimation described above. This is mainly due to the shortages of data on the amount of oils used for the manufacturing and on the amount of oils depleted during the manufacturing, and also due to the technical difficulty of analyses. FSCJ therefore decided to estimate exposures to glycidol derived from such edible oils conservatively focusing on oils in processed

foods, eg, margarine and shortening, on the following assumptions: 1) All of the margarine for business use and a half of the shortening are used for processed foods. There is no loss during the processing. Twenty percent of the fats are discarded during manufacturing, distribution and consumption. 2) The other half of the shortening is used as frying oil for processed foods, and 50% of the frying oil remains in foods. As sums of thus estimated exposures from processed foods and the above-mentioned intakes of edible oils, the average and maximum daily intakes of glycidol derived from GE were estimated to be 1.8×10^{-4} and 2.3×10^{-4} mg/kg bw/day, respectively, and their MOEs of GE were estimated as approximately 9100 and 6900, respectively.

Assuming infants taking only formulated milk powder, the daily intake is estimated to be 7.5×10^{-4} ~1.3×10^{-3} mg/kg bw/day till 5 months after birth, and to be 3.6×10^{-4} ~6.1×10^{-4} mg/kg bw/day for the period from 6 to 11 months after birth. Periods taking formulated milk powder is limited in infancy, and thus it is inappropriate to apply these values for the risk assessment.

Additionally, so far no reports on adverse effects of consuming oils containing GE on human health have been reported. Hence, FSCJ recognized that none of currently available findings suggests adverse effects of GE directly on human health.

5. Conclusion

Adverse effects of edible oils highly containing DAG (hereinafter referred to as DAG oils), which contain GE as contaminants, were not confirmed in animal experiments. Moreover, the tumor promoting activity was not observed in the tests conducted just to confirm it. Potential of tumor promoting activity of DAG oils, once concerned, was thus denied, and FSCJ judged that tumor promoting risk in human from the daily consumption of DAG oils is negligible.

Since the genotoxic carcinogenicity of glycidol derived from GE was not denied, FSCJ has conducted an assessment also on GE and glycidol. The level of GE in the currently available edible oils is maintained as minute, and even if all of the GE were assumed to be converted to the equimoles of glycidol in the body, the extra risks of tumor incidence were estimated to be very low (the exposure levels corresponding to the extra risks of tumor incidence of 10^{-4}, 10^{-5} and 10^{-6} were estimated as 1.6×10^{-3}, 1.6×10^{-4} and 1.6×10^{-5} mg/kg bw/day, respectively), and a certain level of MOE, slightly lower than 10,000, was allocated. Additionally, so far no reports on adverse effects of consuming oils containing GE on human health have not been reported.

While these data suggest no apparent adverse effects due to the consumption of edible oils currently available, the genotoxic carcinogenicity of glycidol was not denied. Therefore, GE exposure levels should be kept as low as possible according to the principle of ALARA (As Low As Reasonably Achievable).

More data on GE such as the toxicokinetics and toxicity on individual GE substances, i.e. to what extent GE are hydrolyzed, absorbed, distributed, metabolized, accumulated, excreted, and consequently exerts toxicity are necessary for future risk assessments. Also, information of human exposure including establishment of methods for analysis of GE in processed foods and the analytical data on the current situation need to be collected. Ingested amounts of edible oils through foods need to be estimated based on total diet studies.