

Original: Japanese
Provisional translation

Evaluation Report of Food Additives

Acetaldehyde

July 2005

Food Safety Commission

Evaluation Results on the Health Risk Assessment of Acetaldehyde as Food Additive

1. Introduction

Acetaldehyde has a fruity aroma, and is naturally contained in foods such as fruits and fruit juice (0.2-230 ppm), vegetables (0.2-400 ppm), dairy products (0.001-76 ppm) and bread (4.2-9.9 ppm) ^{1),2)}. In addition, acetaldehyde is contained in beverages such as tea and soft drinks (0.2-0.6 ppm), beer (0.6-24 ppm), wine (0.7-290 ppm) and spirits (0.5-104 ppm) ²⁾. In Europe and the United States, it is added to reproduce flavors in various processed foods such as soft drinks (average usage: 5.05 ppm) and candy (average usage: hard type: 9.29 ppm, soft type 3.26 ppm) ³⁾.

2. Background

The Ministry of Health, Labour and Welfare, in accordance with the matters approved by the Subcommittee of the Pharmaceutical Affairs and Food Sanitation Council in July 2002, is indicating a policy to start, without waiting for requests of designation from companies and others, a study initiated by the government for the designation of food additives, (1) for which a safety evaluation was completed internationally in the Joint FAO/WHO Expert Committee on Food Additives (JECFA) to ensure their safety to a certain extent, (2) whose use has been approved widely in the U.S., European Union (EU) member states, etc., and for which are considered to have high demands internationally. This time, evaluation data of acetaldehyde as a flavoring agent was collected, and the assessment of the effect of food on health was requested to the Food Safety Commission. (The related documents were received on November 21, 2003.)

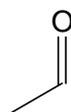
Data on flavors are compiled based not on the "Guidelines for Designation of Food Additives and for Revision of Standards for Use of Food Additives" set out by the Ministry of Health, Labour and Welfare, but on the "Japanese Safety Evaluation Methods for the Flavor Additives Used Widely as well as Internationally".

3. Name, etc.

Name: Acetaldehyde

English name: Acetaldehyde

Structural formula:



Chemical formula: C₂H₄O

Molecular weight: 44.1 g/mol

CAS number: 75-07-0

4. Safety

(1) Genotoxicity

Negative genotoxicity results were reported in the microbial reverse mutation tests ⁴⁾, while positive

results were reported in many test methods using eukaryotes including yeast.

With regard to the test methods using animals, DNA strand breakage by inhalation exposure⁵⁾ is reported, and, in sister chromatid exchange tests using intraperitoneal administration⁵⁾ as well as in micronucleus tests with rodents using intraperitoneal administration⁶⁾ positive results are reported. On the other hand, negative results are reported in micronucleus tests with germ cells by intraperitoneal administration⁵⁾.

In addition, it is reported in one paper that DNA adduct formation was observed in granulocytes and lymphocytes in the blood in a test of DNA adduct formation by acetaldehyde through alcohol intake in humans,⁷⁾ however, it should be noted that this was not a finding that itself indicates mutagenicity

(2) Repeated-dose toxicity

In an 11-week repeated-dose (in drinking water) study (24 male Wistar rats; 0, 120 and 500 mg/kg body weight/day), some degeneration such as that of vesiculate lipid droplets in the liver was observed in the 500 mg/kg body weight/day group, but no effects were observed in the 120 mg/kg body weight/day treated group⁸⁾. The no observed adverse effect levels (NOAEL) was estimated at 120 mg/kg body weight/day.

In a 4-week repeated-dose (in drinking water) study (0, 25, 125 and 625 mg/kg body weight/day) to Wistar rats, kidney weights were significantly increased in male rats in the 625 mg/kg body weight/day administration group. Mucosal hypertrophy of the anterior stomach was observed in the 625 mg/kg body weight/day administration group, and only one female rat among them indicated papillary hyperplasia histologically⁹⁾. The NOAEL was estimated to be 125 mg/kg body weight/day.

(3) Carcinogenicity

Since cancer development was observed in the nasal mucosa in a rat inhalation test (0, 750, 1,500 and 3,000 ppm (dose reduction to 1,000 ppm at 11 months), 6 hours/day, 5 days/week and 27 weeks at the longest) and in the larynx in a hamster inhalation test (dose reduction to 2,500-1,650 ppm, 7 hours/day, 5 days/week, 52 weeks) in the tests carried out by the International Agency for Research on Cancer (IARC)¹⁰⁾, acetaldehyde was classified into Group 2B (possibly carcinogenic to humans)⁵⁾. It should be noted that this was not an oral administration test, conducted to evaluate usual food additives, but an inhalation test.

In a lifetime carcinogenicity test by administering the substance to SD rats in drinking water (50, 250, 500, 1,500 and 2,500 mg/L)¹¹⁾, the cases of developing malignant tumors were increased in the 50 mg/L female and 2,500 mg/L male/female groups. In this test, although the total number of the cases of malignant tumors was increased, the observed tumors were so sporadic that neither dose relationship nor target characteristics was observed, thus, these results were used as reference data for the evaluation of carcinogenesis.

(4) Developmental toxicity

There is no test data using oral administration.

The following reports are available for the tests other than those using oral administration, but they

are used as reference data.

In the developmental toxicity study in rats (8-15 day pregnancy, 50, 75, 100 and 150 mg/kg body weight/day, intraperitoneal injection), the incidence of embryonic death and fetal malformations was increased in all the treated groups.¹²⁾

In the developmental toxicity study in mice (7-9 day pregnancy, approximately 31 and 62 mg/kg body weight/day, intravenous injection), the dose-related increases in incidence of embryonic death and fetal malformations was observed^{10), 13)}.

In the developmental toxicity study by single (6, 7 or 8 day pregnancy) or repeated (6-8 or 7-9 day pregnancy) intravenous injection to mice (0.1 mL of 2% solution/mouse/day), the malformed fetuses were observed^{10), 14)}.

(5) Others

(1) There are no reports in which endocrine disruption was suspected.

(2) Regarding neurotoxicity, it has been identified that protein adduct formation by acetaldehyde through administration of ethanol solution was observed in the brains of infant rats¹⁵⁾. However, the biological effects of protein adduct should merit further research in future, and thus, it is considered that the effects cannot be assessed at this time.¹⁶⁾

5. Estimation of the intake

The estimated daily per capita intake are 19,211 µg and 9,618 µg in U.S. and Europe, respectively, based on the PCTT method of JECFA using a hypothesis that 10% of the population consumes the whole amount of the annual usage of this substance as a flavoring agent^{3), 17)}. Confirmation by a follow-up survey after approval is considered necessary in terms of accuracy, but the estimated intake in Japan can be estimated approximately in the range between 9,618 µg and 19,211 µg since those of the already-approved flavoring agent in Japan are reported comparable to those in the U.S. and Europe. In addition, the volume of intake of this substance as an ingredient contained naturally in food is 4-fold larger than that of this substance added for flavoring purposes¹⁸⁾.

6. Calculation of safety margin

The estimated intake (0.192-0.384 mg/kg weight/day), calculated by dividing the estimated amount of intake (9,618-19,211 µg/person/day) by Japanese average body weight (50 kg), is compared with the NOAEL of 120 mg/kg weight/day in an 11-week repeated-dose toxicity study, and subsequently, a safety margin of 313-625 can be obtained.

7. Evaluation based on structure class

This substance is classified into structure class I since the substance itself and its metabolites are as same as the biogenic substances. The main metabolite is acetic acid, which is further metabolized into carbon dioxide and water which are to be eliminated relatively rapidly into the urine and the breath¹⁷⁾.

8. Evaluation by JECFA

This substance was evaluated by JECFA in 1997 as a group of saturated-fat acyclic branched chain primary alcohol, aldehydes and acids, and was classified into class I. The NOAEL of 125 mg/kg body weight/day (rat) was used. The estimated intake (9,700-11,000 µg/person/day*) exceeds the acceptable intake of class I (1,800 µg/person/day). However, the substance is completely metabolized into biogenic substances, and its level is not predicted to exceed the physiological range; therefore, the substance is considered to be free of any safety concern as a flavoring agent¹⁷⁾.

* Estimated intake used for the evaluation by JECFA

9. Evaluation based on "Japanese Safety Evaluation Methods for the Flavor Additives Used Widely as well as Internationally"

A negative result was obtained in an Ames test on this substance, however, positive results were obtained in other genotoxicity tests; therefore, the substance was considered to have only qualitative genotoxicity. It should be noted that the substance is classified into Class I, that the safety margin (313-625) which is based on an 11-week repeated-dose toxicity study is below the appropriate safety margin (1000), and that the estimated intake (9,618-19,211 µg/person/day) exceeds the acceptable intake of Class I substances (1,800 µg/person/day).

10. Others

Acetaldehyde is extremely soluble in both water and oil and easily absorbed orally, but only minimal amount enters the circulating blood since the majority of it is metabolized by the initial pass effect in the liver or removed by binding to membrane-surface proteins in hepatocytes. Besides, a part of it is also metabolized by aldehyde dehydrogenase (ALDH) within the gastrointestinal tracts including the esophageal mucous membrane, stomach and colon^{8), 19)}. Metabolism by ALDH from acetaldehyde to acetic acid does not generate intermediate metabolites that contain free radicals or substances with other toxicity²⁾, and different routes including a metabolic route via aldehyde oxidase also exist besides that of ALDH²⁰⁾.

ALDH exists in livers and other organs of adults as well as of fetuses and infants^{21), 22), 23), 24)}, and one report indicates that the aldehyde oxidation ability in the livers of human fetuses was equivalent to about 1/10-1/5 of that in adults²⁵⁾.

Although the measurements of the amounts of *in vivo* acetaldehyde synthesis vary widely, some reports state that approximately 1.3 µM²⁶⁾ and 3.9 µM²⁷⁾ of blood acetaldehyde levels can be detected in healthy persons. Though the following values are overestimates, the blood level is not considered to exceed 14 µM even if the estimated daily intake of acetaldehyde in Japan (approximately 19 mg/person/day) is consumed at a time, and if 100% of it is absorbed and distributed in the body without being metabolized by the first pass effect. However, the situation, in which the level of acetaldehyde, equivalent to that used as a flavoring ingredient, is consumed in daily dietary life, is far from being applicable to the above hypothesis. In fact, not all the orally-ingested acetaldehyde is absorbed directly into the body, the majority of it is supposed to change into acetic acid via metabolism by ALDH and other enzymes in the gastrointestinal tracts and liver.

Another report indicates that the metabolic (oxidative) rate of acetaldehyde in nonhuman mammals

is 0.75 $\mu\text{mol}/\text{min}/\text{g}$ liver ²⁾. If the rate in humans is assumed to be comparable to this, the processing capacity in the liver of human adults (approximately 1 kg) is 750 $\mu\text{mol}/\text{min}$ (approximately 33 mg/min); thus, acetaldehyde is considered to be metabolized in the liver within 1 minute even if the intake status is as described above (i.e., approximately 19 mg/day is consumed at a time, and 100% is absorbed), and thus, only minimal amount is considered to enter circulating blood by the first pass effect. In this regard, the maximum level of acetaldehyde in the systemically circulating blood was ≤ 10 μM after approximately 9 mg/kg body weight of acetaldehyde (equivalent to about 24-fold amount of estimated intake (approximately 0.38 mg/kg body weight/day)) was administered into the stomach of male rats ⁸⁾.

In addition, a relationship between the genetic polymorphism of ALDH and alcohol metabolism has been reported. It is known that ALDH II type deficiency is more common among Japanese. Although ALDH II type deficiency is likely to increase the blood aldehyde levels in more susceptible humans than in less susceptible ones, another metabolic pathway is considered to function in a complementary manner ^{20), 28)}.

11. Evaluation results

High-dose inhalation exposure of acetaldehyde has carcinogenicity. A negative result was obtained in an Ames test, while positive results were obtained in other genotoxicity tests. Therefore, this substance is considered to have qualitative genotoxicity, but quantitative evaluation will be needed in the future. Test data on genotoxicity in cancer-target organs has not been obtained.

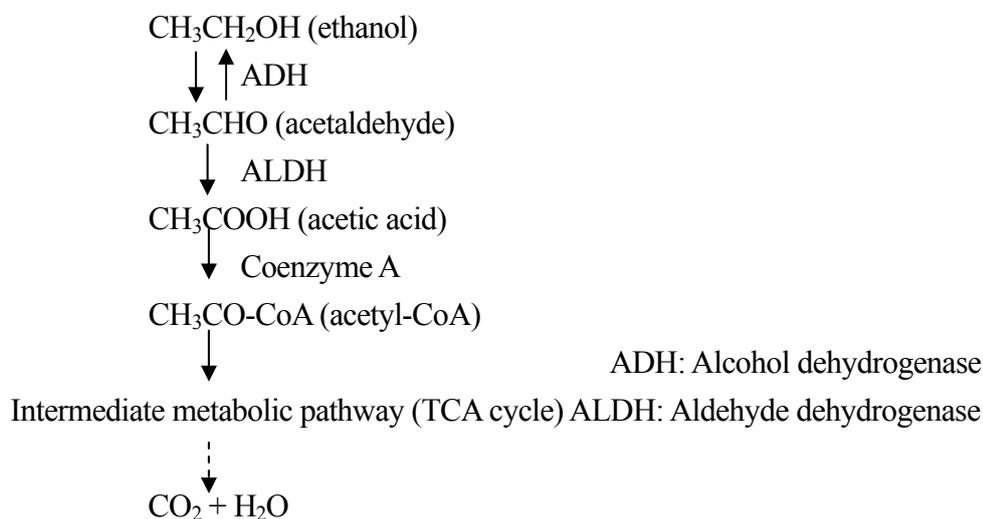
In addition, the estimated intake of this substance exceeded the acceptable intake of class I substances, and the safety margin based on the 11-week repeated-dose toxicity study is below the appropriate safety margin (1,000).

However,

- The dose in the inhalation test is at a higher level than that of the estimated exposure in humans, thus, the observed carcinogenesis was presumed to have resulted from the direct exposure to highly cytotoxic acetaldehyde.
- This substance is consumed from daily foods such as fruit and alcoholic beverages, and the volume of intake is supposed to be larger than that being consumed as flavoring agent.
- The volume presumed to be consumed from food is metabolized by aldehyde dehydrogenase (ALDH) in the gastrointestinal mucosa and eliminated after binding with proteins. In addition, the majority is metabolized by the first pass effect in the liver even if absorbed from the gastrointestinal tract. Therefore, a limited amount is believed to enter the systemically-circulating blood.
- This substance is a biogenic substance, used for a long time in the U.S. and Europe, and no health hazards have been reported due to its use as a flavor.
- According to the evaluation carried out by the JECFA, the substance is classified into class I, and the estimated intake exceeds the acceptable intake of class I, however, this substance is free of safety problems as a flavoring agent because it is presumed to be metabolized completely into biogenic substances and its level do not exceed the physiological range.

By evaluating the above results and reports comprehensively, acetaldehyde is considered to be free of safety concerns when used as a flavoring agent as it is completely metabolized into biogenic substances, and thus, its level is presumed not to exceed the physiological range.

(Reference) Major metabolic pathway of acetaldehyde ²²⁾



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Classification of structures of flavoring ingredients (Acetaldehyde)

YES → NO :→

