

Notification
Letter



Kyowa Hakko U.S.A., Inc.
599 Lexington Avenue, Suite 4103
New York, NY 10022

Telephone: 212 319-5353
Facsimile: 212 421-1283



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Office of Nutritional Products, Labeling and Dietary Supplements
HFS-820
Center for Food Safety and Applied Nutrition
Food and Drug Administration
5100 Paint Branch Pkwy.
College Park, MD 20740
Attention: Dr. Susan Walker, Acting Director of Division of Dietary Supplements

**RE: New Dietary Ingredient Notification
for N-Acetyl-L-Hydroxyproline**

To whom it may concern:

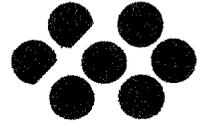
As specified in the Code of Federal Regulations, Chapter 21 CFR, Part 190-Dietary Supplements; Subpart B, Paragraph 190.6 (revised as of April 1, 2002) for Requirement for Premarket Notification we are submitting information about Kyowa Hakko Kogyo Co., Ltd.'s ingredient product **N-Acetyl-L-Hydroxyproline**.

Our intention with this notification is to exhibit to the addressee that **N-Acetyl-L-Hydroxyproline** (hereinafter AHYP), proposed herein as a new ingredient, when used under the suggested intake recommendations and labelings is reasonably expected to be safe. Following the format of the CFR notification requirements for paragraph *(b)* we, Kyowa Hakko U.S.A., Inc., a subsidiary of the manufacturer, supply answers for the requested details as follows.

(1) Name and address of manufacturer: Kyowa Hakko Kogyo Co., Ltd.
Bio-Chemicals Company
1-6-1 Ohtemachi
Chiyoda-ku, Tokyo 100-8185
Japan

Manufacturing site: Kyowa Hakko Kogyo Co., Ltd.
1-1 Kyowa-machi
Hofu-shi, Yamaguchi 747-8522
Japan

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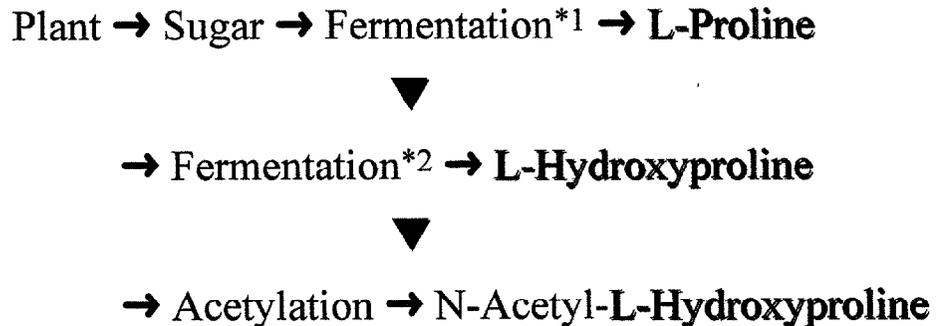
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- (2) **The name of the new dietary ingredient: N-Acetyl-L-Hydroxyproline.**
N-Acetyl-L-Hydroxyproline is a derivative of a natural amino acid, L-Hydroxyproline, which is found in collagen, a most abundant protein in the human body. Kyowa Hakko's L-Hydroxyproline is produced by fermentation and free from potential risk of BSE.

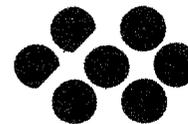
Flow Chart of the production process:



*1: Microorganism

*2: Microorganism producing Proline 4-Hydroxylase (Enzyme)

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(3) A description of the dietary supplement:

- (i) The level of the new dietary ingredient within a supplement will be in the range of 50 mg to 100 mg per tablet or capsule.
- (ii) The conditions of use can be taken daily via oral administration with daily intake of not more than 300 mg with single 50 mg to 100 mg oral doses taken three times per day.

As the agent for the manufacturer of the ingredient, Kyowa Hakko U.S.A., Inc. will responsibly submit the following outline of label directions to our dietary supplement manufacturing and marketing customers.

Statement of identity:

e.g. "N-Acetyl-L-Hydroxyproline"

Net quantity of contents:

e.g. "60 capsules"

Structure-function claim and label wording:

"This statement has not been evaluated by the Food and Drug Administration. This product is not intended to diagnose, treat, cure, or prevent any disease."

"This product is not intended for use by pregnant women and children. The recommended period of taking this product should be no longer than 8 months. Consult with a physician before taking this product for longer durations. If you take any medication, consult with a physician or refrain from taking this product."

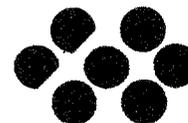
e.g. "A dietary supplement for healthy joints."

Directions for use:

e.g. "Take three capsules daily."

(Daily dosage not to exceed 300mg as N-Acetyl-L-Hydroxyproline)

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- (4) **The history of use and the evidence of safety establish** that N-Acetyl-L-Hydroxyproline (AHYP), when used under the aforementioned conditions is reasonably expected to be safe.

Immediately following herein we summarize information about the safety and the side effects of AHYP reported in animal testing and *in vitro* study. The LD₅₀ data suggest AHYP has no acute toxicity even at a possible erroneous or accidental intake of a high amount of AHYP in a short term. AHYP's non-mutagenicity and non-chromosome aberration activity shown by the *in vitro* study reflect the fact that there has been no report of carcinogenicity from the clinical reports while AHYP has been used in Europe for over twenty years.

The supposed mechanisms of action of AHYP are to help proteoglycan synthesis that is occurring in normal, healthy cartilage.

Safety Data (Animals & In vitro)

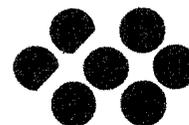
LD₅₀ (oral administration)

7,751 mg/kg of body weight (rats)¹
5,688 mg/kg of body weight (mice)¹
>2,500mg/kg of body weight (rats)¹⁰

29-day administration (4.5; 36; 288 mg/kg of body weight) (rats)¹
No unwanted effects occurred.

28-day administration (4.5; 36; 288 mg/kg of body weight) (dogs)¹
No unwanted effects occurred (4.5; 36mg/kg).
Slight changes on cornea and renal tubules were observed (288 mg/kg).

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Mutagenicity

No indications of any mutagenic potential were found¹.

Ames Test: Negative, up to 5000µg/plate (Maximum dosage examined)¹¹

Chromosome Aberration Test: Negative, up to 1730 µg/ml (Maximum dosage examined)¹²

***Carcinogenicity*¹**

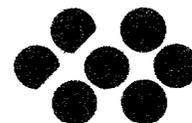
From bioassays and clinical tests, **no indications of a tumorigenic potential** have been reported. **No reproducible teratogenic effects were observed** in rabbits with the dosage of 288 mg/kg of body weight.

***Metabolism/Excretion*⁸**

Ingested AHYP is shown to be completely excreted from the body within a couple of days without being metabolized in dogs.

As exhibited above, an important aspect of short-term and long-term safety of AHYP is that this substance was shown to be unmetabolized by the body. Once AHYP is ingested, it is absorbed and moves to the blood stream. The half-life of AHYP in the blood stream is only several hours and without being metabolized, 100 % of AHYP is excreted from the body. Therefore, AHYP does not accumulate in any part of the body, nor does it produce any deleterious metabolites that should remain in the body for a long term.

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Safety Data (Humans)

600mg/day

24 recipients, 8 weeks

No incidence of non-tolerance²

32 recipients, 6 weeks

No side effects were observed.³

50 recipients, 8 months

No side effects were observed.⁴

100 recipients, 15 days to 6 months.

No incident on any of the cases regarding digestive, hepatic, renal tolerance.⁷

No influence of the cardio-vascular and central or autonomous nervous system.⁷

1200mg/day (2wks) followed by 600mg/day (6wks)

367 recipients

No serious adverse reactions were noticed.

Occasional gastrointestinal complaints (3.54%)⁵.

509 recipients

Low degree side effects were noted (4.7%) mainly on the gastrointestinal tract.⁶

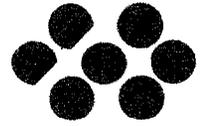
1200mg/day

132 recipients, 21 days

No serious effects were reported.

(18.9% of total: 3 cases of gastrointestinal pain, 3 cases of nausea, 2 cases of skin rash, 2 cases of constipation etc.)⁹

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Open Multicenter Study (600 mg/day)¹³

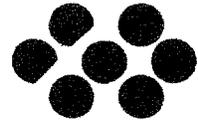
5,523 recipients, 4 and 8 weeks

2.79% (2.3% gastrointestinal complaints, 0.27% allergic reactions, 0.22% others.)**98.8%, very good or good tolerance**

In the clinical study, AHYP is regarded as a well-tolerated and safe substance. However, as is reported in the reference # 9, the percentage of side effect (SE) occurrence at the dosage of 1200mg/day is not negligible. The SE occurrence can be judged as dose dependent between 600mg/day and 1200mg/day, judging from the literature. At the generally prescribed dosage of 600mg/day, SE is thought to occur none^{2,3,4,7} to 4.7%⁶, while at 1200mg/day the figure is expected to be 18.9%⁹, most of which should be from temporary gastrointestinal complaints. From the comparison of the SE frequencies between the two intake levels (600mg/day and 1200mg/day), determining the recommended intake of AHYP to 300mg/day assures the significant reduction of SE frequency to a back ground level.

Although reference # 13 showed 2.79% SE in 8 week study, references # 4 and # 7 reported no side effects in 8 months and 6 months studies, respectively, suggesting that the liability of SE does not increase for over 8 weeks of AHYP intake. In other words, if the SE frequency were dependent on intake duration for over 8 weeks, the studies in references # 4 and # 7 should have resulted in significantly higher SE frequency than 2.79% (reference # 13), but it was not the case. This led us to conclude, in addition to limiting the daily intake of AHYP to 300mg/day, that fixing the recommended duration of AHYP intake to 8 months must make it possible to avoid any possible SEs that occur by taking AHYP supplements for a period longer than described in scientific literatures.

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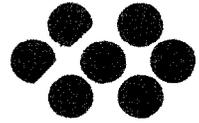
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Based on these views, we determined AHYP's use as an ingredient for dietary supplements as follows. These instructions will provide significant guidelines to reinforce the safe use of AHYP within dietary supplements.

- I. AHYP is intended to be used as an ingredient of dietary supplements that help maintain healthy joints and is not to be used by those who have medication of arthritis from doctors.
- II. The recommended intake of AHYP is limited to 300mg/day. Each label of the product including AHYP shows the recommended dosage of the product based on the amount of AHYP to be taken in a day.
- III. Dietary Supplement labels of the product including AHYP are to show "This product is not intended for use by pregnant women and children" "The recommended period of taking this product should be no longer than 8 months. Consult with a physician before taking this product for longer durations."
- IV. Each label of the product including AHYP shows "If you take any medication, consult with a physician or refrain from taking this product."

The above four (IV) points reiterate our labeling instructions outlined on page 3 of this document.

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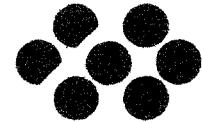
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**(5) The signature of the person designated by the manufacturer
(Kyowa Hakko Kogyo Co., Ltd.) of the ingredient is:**

Neil C. Sullivan
Manager
Kyowa Hakko U.S.A., Inc.

Thank you in advance for your attention and considerations in this matter. We look forward to receiving acknowledgment from the FDA that this notification has been duly received.

In the event you need to contact Kyowa Hakko, please contact the designee noted above.

Sincerely,

Toshifumi Asada
CEO and President
Kyowa Hakko U.S.A., Inc.

Enclosures