New Technology/Ingredient Preliminary Assessment Form

The intended use of this form is to provide suppliers, and potential partners, guidance and direction for technology selection and evaluation. After receiving the Preliminary Assessment Form, New Technology R&D staff should be able to evaluate submissions more efficiently and streamline the decision making process.
ONLY share NON-CONFIDENTIAL information until a CDA is in place

Supplier and Ingredient Details

Please provide the following information

PROJECT NAME (Ingredient Trade Name): HEPA-L1 Project(Agrimony Extract) **Active Ingredient Scientific Name: Agrimonia eupatoria** **Submission Date: April 22. 2010**
Description of Product: Agrimony is food material improving alcoholic liver function and antioxidant.
Supplier Company Name: BIOKOREA CO., Ltd. Contact Name: **Lee Minkyung** Address: #807, Mario Tower, 222-12, Guro-Gu, City, Country: Seoul, 152-050, KOREA E-mail Address: parrot76@empal.com Phone number: +82-2-538-8825 Fax number: + 82-2-890-0545 Company Website: www.liverkorea.com("Healthkhan® " site)
Manufacturer Name: (if ingredient is contract manufactured or not manufactured by the Company): Contact Name: Address: City, State: E-mail Address: Phone number: Fax number:

Technology Submission

1. **Supporting Evidence (Efficacy/Functionality):**
   Provide all supporting literature and research studies, publications, protocols, and analytical assessment.
   Please separate data into:
   - Mechanism of Action/s (MOA)
     : Non-Nucleoside Anti-viral fraction of aqueous extract of *Agrimony* that interferes with enzymes in viral replication
   - Chemistry/Characterization data ; n.a.
   - *In vitro* bioassay data

     Inhibition of Hepatitis B Virus by an Aqueous Extract of *Agrimonia eupatoria* L.

     An aqueous extract of ***Agrimonia*** was confirmed that it showed a strong suppressive effect on the expression of the hepatitis surface antigen (HBsAg) and hepatitis e antigen

(HBeAg) from the HepG2.2.15 cell line transfected with hepatitis B virus DNA (HBV DNA), and have a slight inhibitory effect on the cell viability of cells for 2days. Glycyrrhizin showed strong inhibition effect on the expression of HBsAg, but have little effect on inhibition of HBeAg, and lamivudine have little inhibition effect for HBsAg and HBeAg for 2days. HBV DNA in the medium of the HepG2.2.15 cells treated with the aqueous extract of Agrimonies was decreased as a dose dependent manner, and the aqueous extract of Agrimonies inhibited the HBV polymerase activity. The reducing effect of HBsAg in transgenic mice orally administrated with the aqueous extract of Agrimonies (300mg/kg per day) was effective as compared to placebo-treated controls. 120 ICR mice were orally administrated with the extract; LD50 of the extract was 1.72g/kg. In this result, the aqueous extract of Agrimonies showed significant inhibitory activity on HBsAg and HBV DNA released from HepG2.2.15 cells. And this transgenic mouse model was similar to what would be predicted from treatment of HBV infected human patients with Agrimonies. ***PHYTOTHERAPY Research 2005; 19(4):355-8 (ISSN: 0951-418X)***

- Inhibition effect of HBsAg in serum before and after treatment of A. extract.

Time	Placebo group(%)	*Agrimonies Extract*	
		300mg/kg/day	1000mg/kg/day
Day 0	100	100	100
Day 7	146.75±46.35	37.12±33.12	21.37±15.29

* The inhibition of HBsAg was analysed by radioimmuniassay (RIA) and the inhibition percentage was calculated using the following equation: HBsAg(%)=[HBsAg at day 0/ HBsAg at day 7]x 100. Values are mean±SD for three mice.

○ *In vivo* testing (Laboratory, Animal, and Human Clinical Studies)

Hepatoprotective Effects of Aqueous Extract from Aerial part of Agrimmony

Hepatoprotective effects of an aqueous extract prepared from the aerial parts of ***Agrimonia eupatoria*** L., a species of agrimmony, were investigated in experimental liver-damaged models. To investigate hepatoprotective effects, the agrimmony extract were fed orally to experimental animals. Thereafter a single dose of hepatotoxin, carbon tetrachloride (CCl_4) or D-galactosamine was orally administrated. Chronic liver damage was induced by oral administration of CCl_4 for 2 weeks (1 time / day). Hepatoprotective effects were monitored by estimating serum AST and ALT levels. The results showed that the agrimmony extract significantly reduced AST and ALT levels compared with those of control group in both acute and chronic animal models. It was concluded that the agrimmony extract have hepatoprotective effects against rat liver injury induced by CCl_4 or D-galactosamine. Ko. J. Pharamacogn***, 2006, 37(1) : 28~32***

Table 1. Body and liver weights of male SD rats orally treated with CCl_4 and *Agrimonia eupatoria*

Group	VC	CON(-)	CON(+)	Low dose (16mg/kg)	Middle dose (80mg/kg)	High dose (400mg/kg)
B.W.(g)	212.7±9.8	216.1±4.6	213.3±7.6	209.2±4.4	209.0±6.1	210.6±5.2
Liver Weight(g)	9.2±1.3	9.7±0.7	10.4±1.0	9.5±0.6	9.5±0.4	9.7±0.6

VC; Olive oil, CON(-);CCL_4, CON(+); CCL_4+UDCA, Silymarin 25mg/kg; Treatment groups; CCL_4+Agrimonia eupatoria Extracts.

Table 2. Hematologyical and serum biochemical values of male SD rats orally treated with CCl_4 and Agrimonia eupatoria

Group	VC	CON(-)	CON(+)	Low dose (16mg/kg)	Middle dose (80mg/kg)	High dose (400mg/kg)
GOT	193.0±30.0	349.7±19.2	256.3±12.3**	221.3±43.3**	214.3±56.9**	385.0±95.5
GPT	48.2±8.7	178.0±15.0	124.8±6.7**	125.2±12.8**	95.2±23.6**	138.3±50.3
LDH	2530.2±262.5	2785.2±947.5	2864.2±541.3	2251.5±741.5	1918.8±583.6	2514.7±670.1
WBC	9.63±1.44	9.48±1.10	9.48±1.40	11.32±1.77	10.06±0.80	9.48±1.71
RBC	6.61±0.14	6.46±0.52	6.77±0.39	6.86±0.25	6.42±0.22	6.81±0.25
Hb	14.70±0.30	14.40±0.60	14.70±0.58	15.23±0.32	14.42±0.37	14.82±0.34
HCT	43.03±1.11	41.52±1.89	42.24±1.80	44.22±1.39	41.28±1.20	42.98±1.03

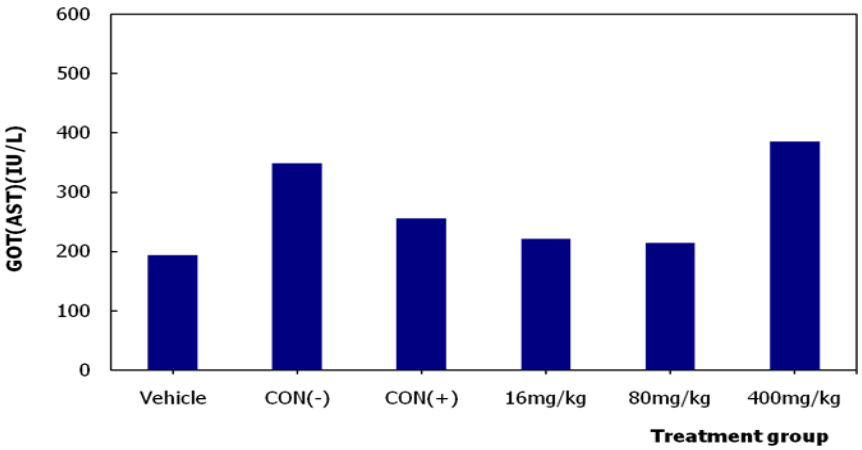


Fig. 1. Serum AST levels.

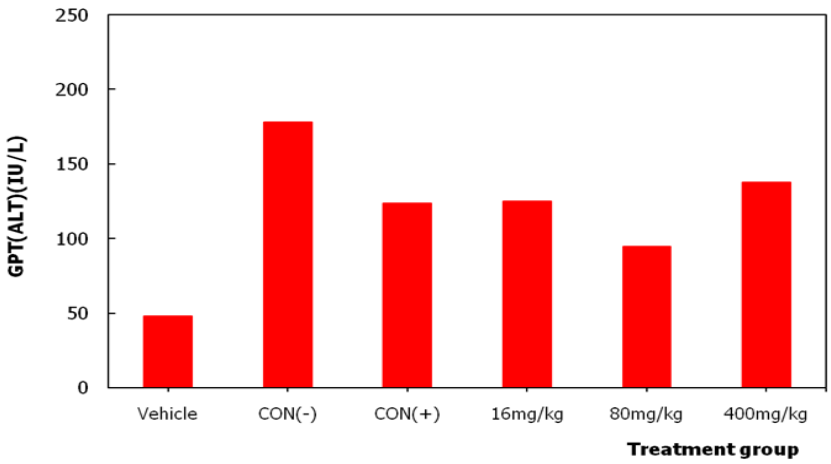


Fig. 2. Serum ALT levels.

Table 1. Body and liver weights of male SD rats orally treated with D-galactosamine/LPS and Agrimonia eupatoria

Group	VC	CON(-)	CON(+)	Low dose (16mg/kg)	Middle dose (80mg/kg)	High dose (400mg/kg)
B.W.(g)	212.7±9.8	207.4±2.4	210.1±4.7	214.6±5.2	210.7±8.4	212.2±7.9
Liver Weight(g)	9.2±1.3	6.3±0.4	7.1±0.9	7.6±0.6	7.9±0.1	7.9±1.5

VC; Olive oil, CON(-);CCL_4, CON(+);CCL_4+UDCA, Silymarin 25mg/kg; Treatment groups; CCL_4+Agrimonia eupatoria Extracts.

Table 2. Hematologyical and serum biochemical values of male SD rats orally treated with D-galactosamine/LPS and Agrimonia eupatoria

Group	VC	CON(-)	CON(+)	Low dose (16mg/kg)	Middle dose (80mg/kg)	High dose (400mg/kg)
GOT	203.5±15.0	1261.2±99.4	337.4±112.3**	263.0±37.7**	220.2±25.4**	269.4±45.2
GPT	51.2±4.5	675.4±6.8	263.4±24.2**	176.4±88.6**	89.2±20.6**	167.0±53.5
LDH	2535.6±293.1	919.0±303.6	821.4±159.5	1035.3±137.7	1046.8±155.4	768.0±463.4
WBC	9.65±1.86	16.46±4.59	11.90±2.33	13.02±3.70	11.13±4.41	9.12±4.19
RBC	6.62±0.18	8.13±0.45	7.94±0.40	7.52±0.25	7.16±0.21	7.59±0.59
Hb	14.70±0.39	17.72±0.77	17.60±0.80	16.56±0.88	16.03±0.29	16.50±1.07
HCT	43.05±1.44	51.92±3.09	51.04±2.75	48.18±2.45	46.30±0.83	47.64±3.22

- Clinically tested : plan to conduct clinical trial in 2010
  - Provide studies done by Company on the ingredient.
  - Total number of clinical studies completed?
  - Study design? (number of subjects, duration of the study/ies, double-blinded, placebo-controlled, case controlled, etc.)
  - Percent response rate?
  - Dose and delivery form used in the study/ies

- Published or not published, and in which journal/s?
  - The Korea society of Gatroenterology (2001.11.23.SheratonWalkerhill) Antiviral Activities of an Extract from Agrimonia eupatoria L. on Hepatitis B Virus:invitro and invivo studies
  - PHYTOTHERAPY Research 2005; 19(4):355-8 (ISSN: 0951-418X) Inhibition of hepatitis B virus by an aqueous extract of Agrimonia eupatoria L.
  - The korean Society of Pharmacognosy Kor.J.Pharmacon 37(1) 28~32(2006) Hepato protective Effects of Aqueous Extract from Aerial part of Agrimony In vivo
  - The Pharmaceutical Society of Korea Arch Pharm Res Vol 27, No 9, 944-946, 2004 An isocoumarin with hepatoprotective activity in HepG2 and primary hepatocytes from Agrimonia pilosa.

- Recommended Delivery form/s?
  : Flexible delivery forms such as tablet, hard & soft capsule, granule and liquid

- Recommended dose/s? (mg per day)
  In case of HEPA-L1(Brand name of Agrimony Extract)
  - Dried Herb: 1~4 gms tds
  - Infusion: 40~80 mls tds
  - Fluid Extract: 2~3 mls tds
  - Tincture: 1~3 mls tds

- Collaborating Organizations, professors or University Affiliations
  - Korea Research Institute of Bioscience Biotechnology
  - Gyeonggi Pharmaceutical Research Center
  - Prof. Okpyo-Zee, The College of Pharmacy, Sungkyunkwan University
  - Prof. Seoung Kew Yoon, The Catholic University of Korea, School of Medicine, the department of Gastroenterology at Seoul St. Mary`s Hospital
  - Prof. Yung Ho, Lim , Sructural Biological Chemistry Lab. Konkuk University
- Conclusions from the studies

2. Intellectual Property / Exclusivity

Provide an IP Portfolio summary to include:

- Provide patent information
  - Provisional/non-provisional/PCT/
  - How is the patent unique, compared to competition?
  - Number of patents? 5
  - Patent Type? (Registration)
  - Patent Number/s, title, abstract, claims and application/issue dates

Registered No.	Title	Exclusivity	Application date
10-2000-003489	Novel extract from *Agrimonia pilosa L.* inhibiting synthesis of surface antigen of hepaptitis B virus, process for preparation the same and the use thereof	Korea	00.01.25
10-2000-0015152	B type viral hepatitis HBsAg control extract isolated from *Lepidium apetalum W.* and process for isolation thereof	Korea	00.03.24
10-2002-0061482	Methods for producing Agrimonia extract with improved activity against Hepatitis B Virus and pharmaceutical and food compositions containing said extract	Korea	02.10.09
CN1466462A	Methods for producing Agrimonia extract with improved activity against Hepatitis B Virus and pharmaceutical and food compositions containing said extract	China	02.10.09

- Describe exclusivity options (MLM, all markets, global, etc.)
  - Korea : Supply raw materials(All markets)
  - China : Supply raw materials & Products(All markets)

3. Safety & Regulatory

- Specification sheet/Technical Data Sheet

Can be provided with CDA. Below is a COS containing appearance, water content, and *E.coli*

2009- 1 -23 02:19 From:F&B BIO 0415558301 To:028900545 Page:1/1
'09-01-23 15:39 FROM- T-095 P001/001 F-932

시 험 성 적 서

검사자	확인자

귀 중

품 명	아그리모니 농축 분말	Lot No.	09012101
		발 행 번 호	09 - 01 - 23
제 조 일 자	2009년 01월 21일	시 험 일 자	2009년 01월 22일
성 상	고유의 색택을 지니며, 짙은나름 특유의 맛과 향취.		

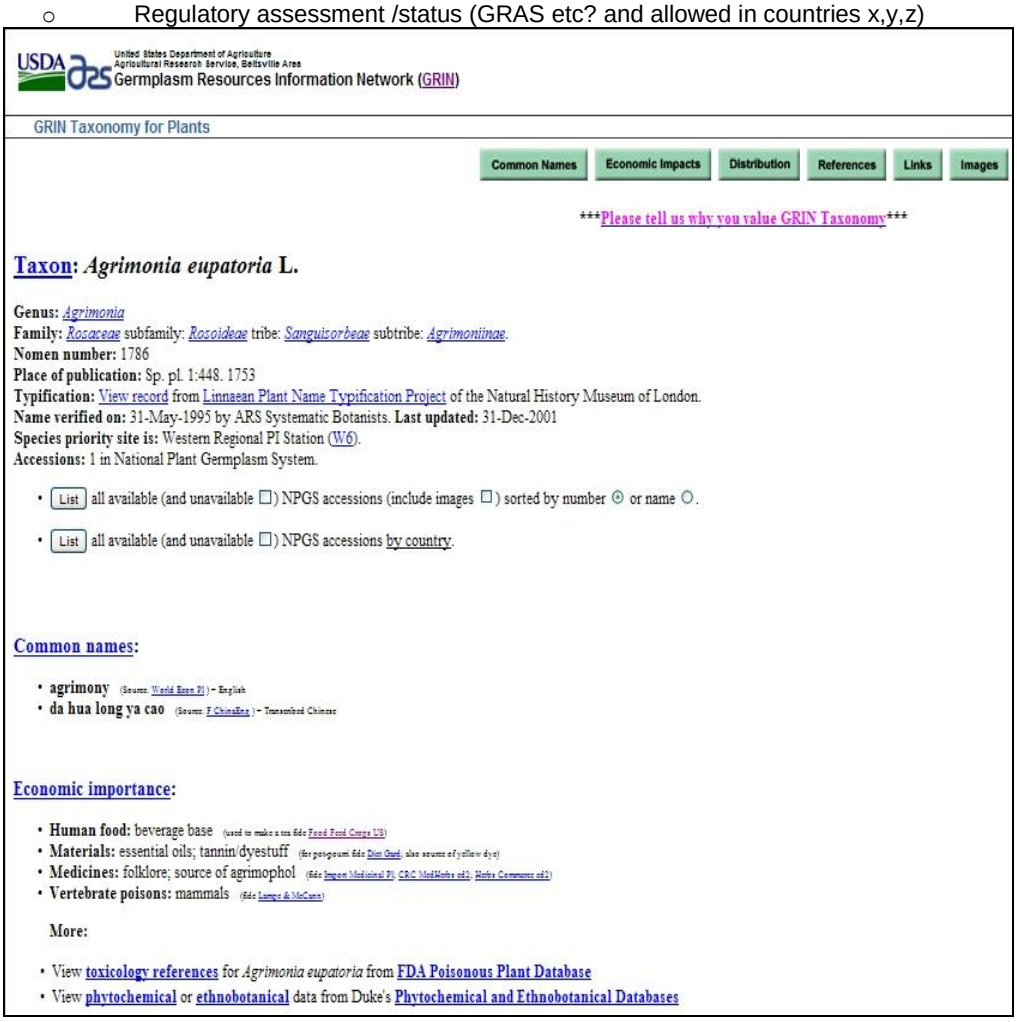
	시 험 항 목	unit	규 격	분 석 결 과	분 석 방 법	비 고
1	성 상			적 합	SENSORY TEST	
2	수 분	%	7% 이하	4.8%	건조 감량법	
3	대 장 균		음 성	음 성	식품공전 시험법	
4	이 물		돌 검 출	불검출	관능검사	
5						
6						
7						
8						
시 험 법	※ 상기 분석결과는 당사 품질관리팀에서 분석한 결과임을 통보합니다.					

(주)코 시 스 품 질 관 리 팀

종 합 판 정

서울사무소 : 서울시 구로구 구로동 187-10 코오롱싸이언스밸리 1차 11층
TEL : 02) 865-8887 FAX : 02) 858-9191
본 사 : 충남 천안시 수신면 백자리 384-16
TEL : 041) 552-1151-2 FAX : 041) 552-1153

- Regulatory assessment /status (GRAS etc? and allowed in countries x,y,z)



United States Department of Agriculture
Agricultural Research Service, Beltsville Area
Germplasm Resources Information Network (GRIN)

GRIN Taxonomy for Plants

Common Names | Economic Impacts | Distribution | References | Links | Images

Please tell us why you value GRIN Taxonomy

Taxon: *Agrimonia eupatoria* L.

Genus: *Agrimonia*
Family: *Rosaceae* subfamily: *Rosoideae* tribe: *Sanguisorbeae* subtribe: *Agrimoniinae*.
Nomen number: 1786
Place of publication: Sp. pl. 1:448. 1753
Typification: View record from Linnaean Plant Name Typification Project of the Natural History Museum of London.
Name verified on: 31-May-1995 by ARS Systematic Botanists. **Last updated:** 31-Dec-2001
Species priority site is: Western Regional PI Station (W6).
Accessions: 1 in National Plant Germplasm System.

- List all available (and unavailable ☐) NPGS accessions (include images ☐) sorted by number ◉ or name ○.
- List all available (and unavailable ☐) NPGS accessions by country.

Common names:

- **agrimony** (Source: World Econ PI) – English
- **da hua long ya cao** (Source: F ChinaFlora) – Transcribed Chinese

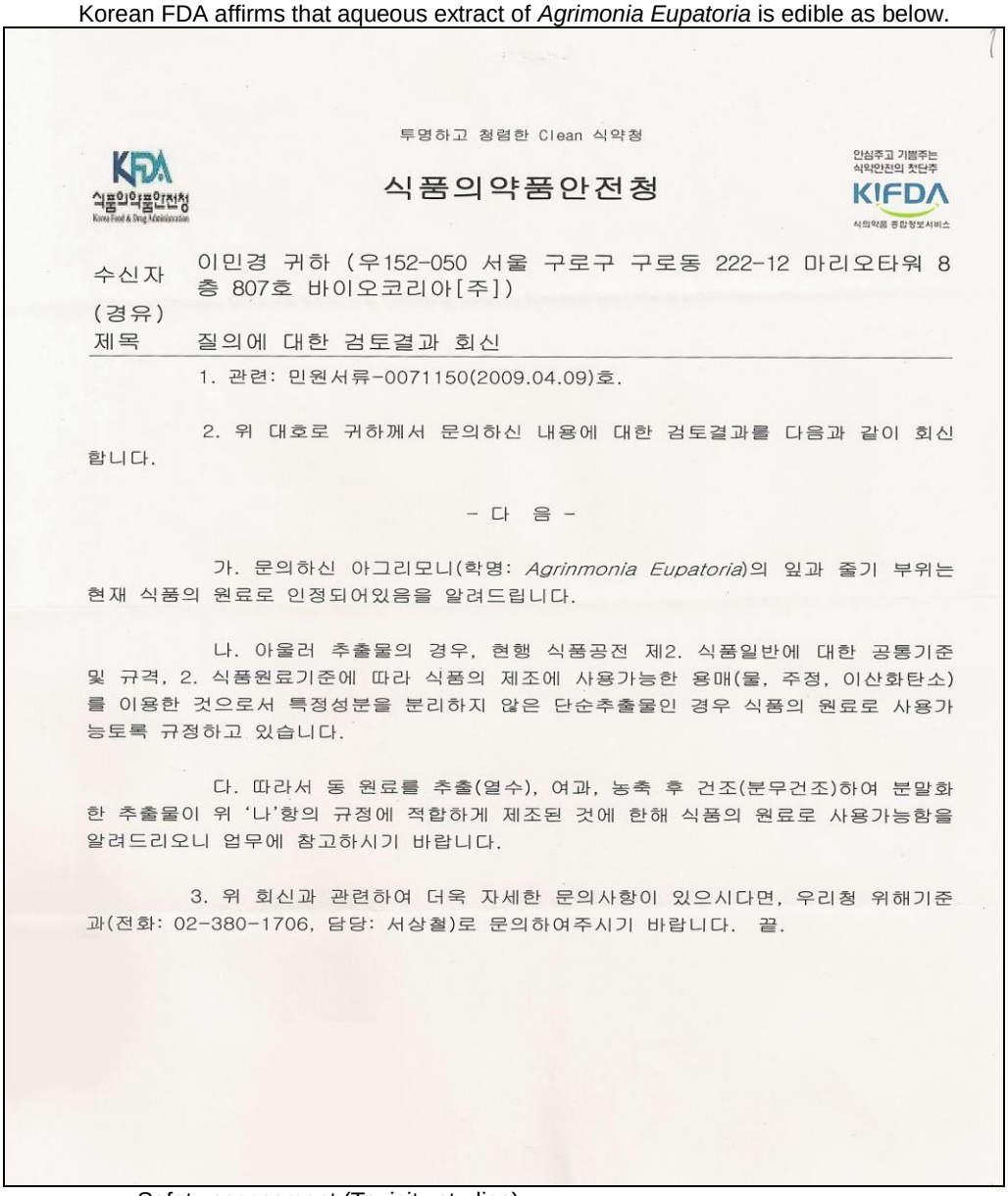
Economic importance:

- **Human food:** beverage base (used to make a tea fide Food Food Crops US)
- **Materials:** essential oils; tannin/dyestuff (for polygonum fide Dict Gard; also source of yellow dye)
- **Medicines:** folklore; source of agrimophol (fide Ingram Medicinal Pl; CRC MedHerbs ed2; Herbs Commerce ed2)
- **Vertebrate poisons:** mammals (fide Lampe & McCann)

More:

- View toxicology references for *Agrimonia eupatoria* from FDA Poisonous Plant Database
- View phytochemical or ethnobotanical data from Duke's Phytochemical and Ethnobotanical Databases

Korean FDA affirms that aqueous extract of *Agrimonia Eupatoria* is edible as below.



투명하고 청렴한 Clean 식약청

식품의약품안전청

안심주고 기쁨주는 식약안전의 첫단추 KIFDA 식의약품 종합정보서비스

수신자 이민경 귀하 (우152-050 서울 구로구 구로동 222-12 마리오타워 8층 807호 바이오코리아[주])

(경유)

제목 질의에 대한 검토결과 회신

1. 관련: 민원서류-0071150(2009.04.09)호.

2. 위 대호로 귀하께서 문의하신 내용에 대한 검토결과를 다음과 같이 회신합니다.

- 다 음 -

가. 문의하신 아그리모니(학명: *Agrinmonia Eupatoria*)의 잎과 줄기 부위는 현재 식품의 원료로 인정되어있음을 알려드립니다.

나. 아울러 추출물의 경우, 현행 식품공전 제2. 식품일반에 대한 공통기준 및 규격, 2. 식품원료기준에 따라 식품의 제조에 사용가능한 용매(물, 주정, 이산화탄소)를 이용한 것으로서 특정성분을 분리하지 않은 단순추출물인 경우 식품의 원료로 사용가능토록 규정하고 있습니다.

다. 따라서 동 원료를 추출(열수), 여과, 농축 후 건조(분무건조)하여 분말화한 추출물이 위 '나'항의 규정에 적합하게 제조된 것에 한해 식품의 원료로 사용가능함을 알려드리오니 업무에 참고하시기 바랍니다.

3. 위 회신과 관련하여 더욱 자세한 문의사항이 있으시다면, 우리청 위해기준과(전화: 02-380-1706, 담당: 서상철)로 문의하여주시기 바랍니다. 끝.

- Safety assessment (Toxicity studies)
  - Safety established *in-vitro*
  - Safety established *in-vivo*

Toxicity of *Agrimonia E.* extract by oral or intraperitoneal(i.p) administration in ICR mice

	Only vehicle	Oral administration		i.p.administration		
		0.125g/kg	0.25g/kg	0.5g/kg	1g/kg	2g/kg
Extract of Agrimonies	0/7	0/7	0/7	1/7	6/7	7/7

* Value of LD 50 represented is 1.72g/kg/day at i.p. administration

- MSDS sheet : n.a.

- Certificate of Analysis (C of A)

No.	Concentration of test liquid		Contents of original (%)		Contents of Extracts (%)	
	㎍/mL	Average (㎍/mL)	%	Average (%)	%	Average (%)
1	0.82	0.71	0.0033	0.0029	0.026	0.022
2	0.73		0.0029		0.023	
3	0.59		0.0024		0.019	

- GMO Status or Non-GMO Statement : Non GMO
- Product promotional marketing literature
- Pesticide data : passed the kFDA requirement s as below.

Pesticide	Less than (mg/kg)
BHC(total of *α,β,γ* and *δ*-BHC)	0.2
DDT(total of *p,p*'-DDD, *p,p*'-DDE, *o,p*'-DDT and *p,p*'-DDT)	0.1
Aldrin	0.01
Endrin	0.01
Dieldrin	0.01

- Heavy metals testing sheet
  : passed the KFDA requirement(less than 30mg/kg of total heavy metals)
- Storage information : RT at dark room with sealed
- Stability Test studies and results : under preparation
- Country/ies of Origin : Korea
- Plant Part/s Utilized (seed, root, skin, rhizome, bark, bulb, whole fruit, leaves, flower) : Whole plant except root
- Variety or cultivar (plant source genus and species) : *Agrimonia Eupatoria*
- Methods of Analysis (or Reference Standard to identify species)
- Extraction Method/s (brief description of extraction process)
  - Are solvents used in extraction? No. Just aqueous extraction
  - Which solvents utilized? Just aqueous extraction
  - Residual Solvent Level (ppm)
  - Final Extraction Ratio (crude botanical: finished product)
    : varied depending on the final conc.
- Source of Raw Material (Natural, Synthetic, Animal, Biofermentation, or describe other) : Natural
- Product Specification : available
- Kosher/halal Certification and expiration date?: n.a.
- Is botanical Certified Organic? : not yet
- Statement of non-irradiation : non-irradiation
- Nutrient Profile
  : bitter principles, Essential oils, Iron, Nicotinic acid, silica, silicic acid, Tannins, Vitamins B & K
- Manufacturing flow chart
  Raw material -> washing -> aqueous extraction : 2 times -> filtration -> concentration -> mixing with dextrin -> SD -> final product

4. Market Assessment

- Marketing Assessment
  - Market size (Top Five Target Markets)
    The potential market of Korea consists of liver improvement(OTC : 150mil US$, health food : 100mil US$) and anti-hangover drink(100mil US$).
  - Consumer studies/interest
    Currently our partner is conducting clinical trial for anti-hangover drink and interim result shows superior efficacy against current competitors.
- Potential Claims and /or approved Health Claims and Benefits
  - Improvement of ALT and AST
  - Anti-Hepatitis
  - Improvement of alcohol induced liver damage
  - Anti-hangover
- Competitive Review
  Patent-protected material with various claims compared to competitors such as Hovenia Dulcis Fruit, Mushroom Mycelia and Milk Thistle. Therefore we have strong position in Market.
- Technology Comparison

products	Patent	claims
Extract of Agrimony	Registered in Korea	Acute & chronic liver disease Anti hepatitis
Extract of Hovenia Dulcis Fruit	No patent	Alcohol induced liver damage
Extract of Mushroom Mycelia	expired	General liver improvement
Extract of Milk Thistle	expired	General liver improvement

- Other Marketing Research and Analysis

5. Commercial Viability

To commercialize this product, please describe your production capability.

- Material cost (per kilo/#/gm/IU/mcg/mg) : 1,000,000 KRW/Kg (as Powder form)
- Forecast

  The major liver disease market is Asia which covers over 75% of worldwide market. Korea and China especially have huge potential market of alcohol induced liver damage as health food. Considering current efficacy data of Agrimony, we strongly believe that this product can meet the market needs and also establish strong market position easily because of intellectual protection in Korea and China as well as efficacy.
- Supply and inventory available in large scale, and at what quantity?

  : We have our own farm in Gyeongbuk(Korea) area as well as China.