

Procyanidin B-2 and the Hair-Growing Activity of Proanthocyanidins

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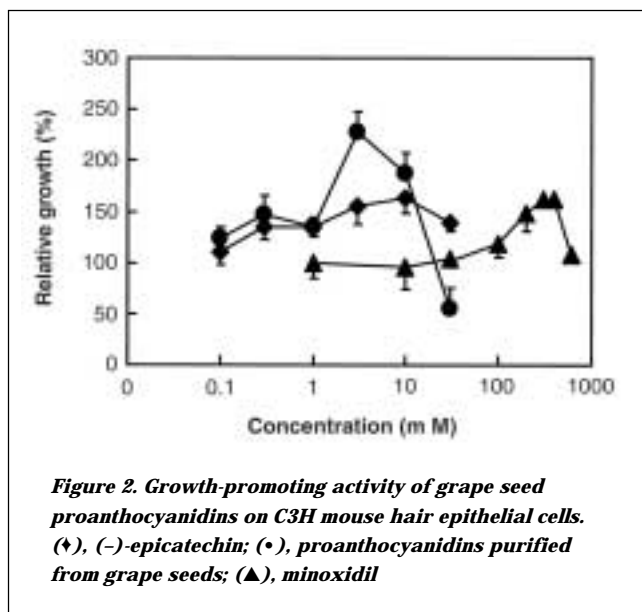
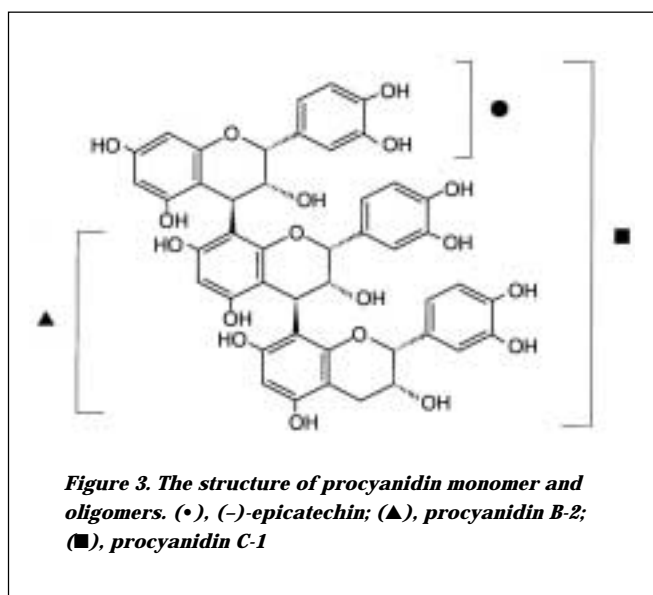
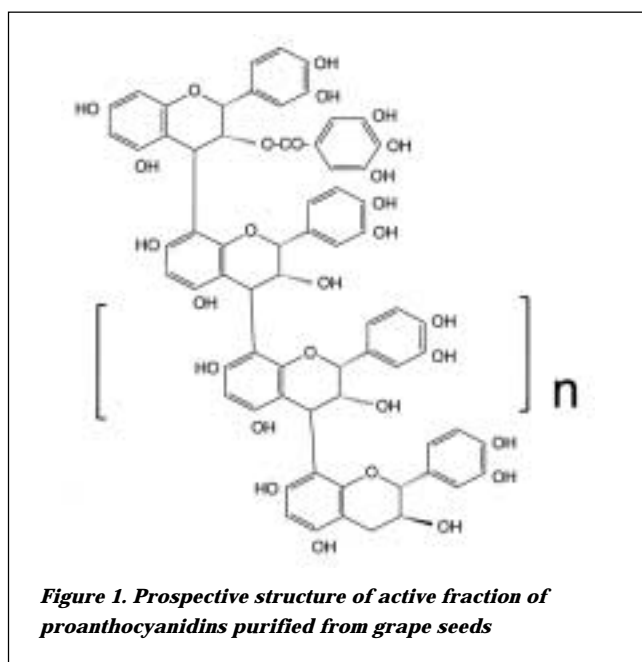


Table 1. Characteristics of the active fraction of proanthocyanidins purified from grape seeds

	Terminal unit	Extension units
Catechin	1	2.3
Epicatechin	2.8	8.1
Catechin gallate	-	-
Epicatechin gallate	1.3	3.4
Catechin : epicatechin	1 : 4.7	
Degree of polymerization	3.7	
Extent of galloylation	25%	

Table 2. Toxicological studies on procyanidin B-2

Test item	Organism	Test substance	Results
Mutagenicity			
Reverse mutation	<i>S. typhimurium, E. coli</i>	PB2*	non-mutagenic
Chromosomal aberration	CHL cells	PB2	no structural aberration
Micronucleus	mouse	PB2	negative
Primary irritation	rabbit	Preparation	no primary irritation
Acute subcutaneous injection	rat	PB2	LD ₅₀ * > 2000 mg/kg
Skin sensitization	guinea pig	PB2	extremely low
Primary ocular irritation	rabbit	Preparation	slight irritation**

*PB2 = Procyanidin B-2
** = same as placebo

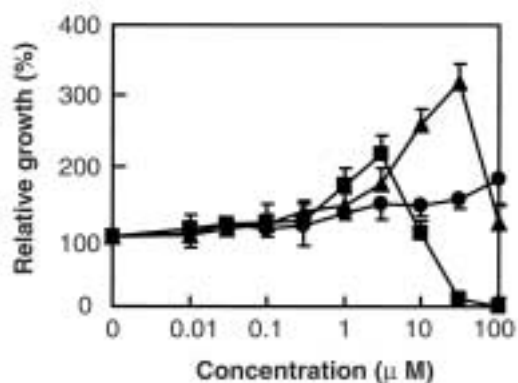


Figure 4. Growth-promoting activity on C3H mouse hair epithelial cells by procyanidins of different degree of polymerization. (•), (-)-epicatechin; (▲), procyanidin B-2; (■), procyanidin C-1

Formula 1. Topical solution for the in vivo mice test

Ethanol	70.00% (w/w)
Procyanidins* or minoxidil	1.00
1,3-Butylene glycol	10.00
N-Acetylglutamine isostearyl ester (Kyowa Hakko Kagyo Co.)	0.50
Polyoxyethylene (25) glyceryl monopyroglutamate monoisostearate (Nihon Emulsion Co.)	0.25
Water (<i>aqua</i>), purified	18.25
	<hr/> 100.00

* Either procyanidin B-2 or procyanidin C-1

Table 3. Background factors

Group	Number of subjects	Mean age	Patterns of baldness ^a		Degrees of baldness ^b		
			Type II	Type IV	Slight	Moderate	Severe
Placebo	10	48	6	4	2	6	2
PB2*	19	45	11	8	4	11	4

^aAccording to the Ogata scale^{23,24}

^bPlaced in three ranks: slight, moderate, and severe

*PB2 = procyanidin B-2

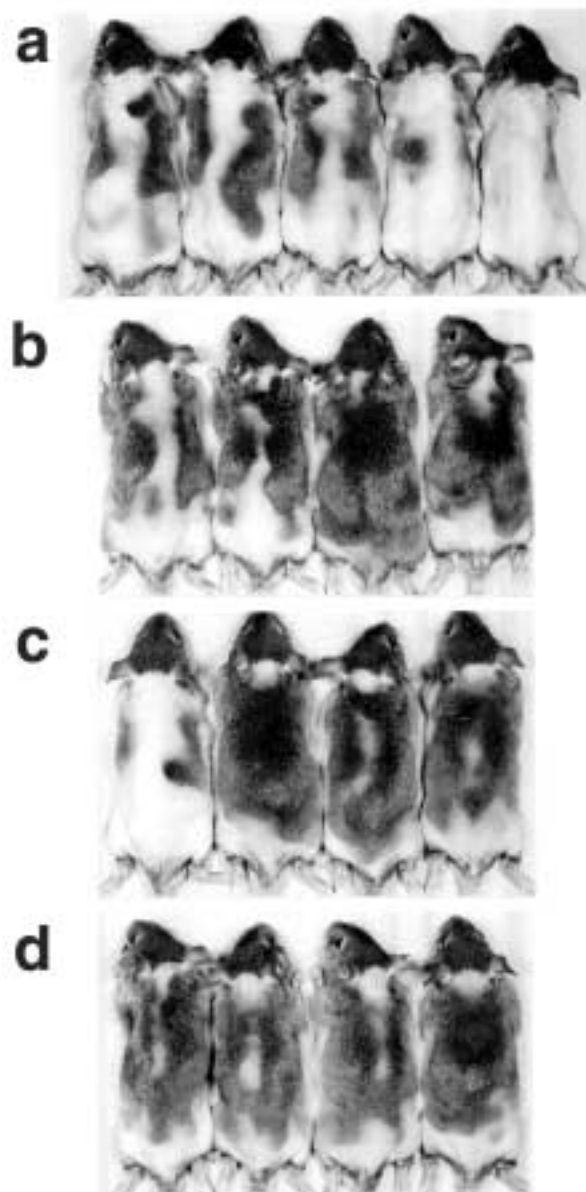


Figure 5. Hair-growing activity evaluated using the C3H mouse in vivo model. (a), vehicle; (b), 1% minoxidil; (c), 1% procyanidin B-2; (d), 1% procyanidin C-1

Formula 2. Hair tonic for the clinical trial

Ethanol	70.00% (w/w)
Procyanidin B-2	1.00
1,3-Butylene glycol	10.00
N-Acetylglutamine isostearyl ester (Kyowa Hakko Kogyo Co.)	0.50
Polyoxyethylene (25) glyceryl monopyroglutamate monoisostearate (Nihon Emulsion Co.)	0.25
<i>d</i> l- α -Tocopherol	0.10
<i>d</i> -Biotin	0.05
Ascorbyl palmitate	0.10
β -Carotene	0.001
Sodium citrate	0.10
Water (<i>aqua</i>), purified	17.899
	100.00

Role of the Outer Root Sheath and the Hair Bulb

Hair is composed of epithelial cells, such as inner root sheath cells, outer root sheath cells and hair matrix cells, and mesenchymal cells such as dermal papilla cells.

Interactions between epithelial cells and mesenchymal cells are considered to be important in the progress of the hair cycle through the anagen, catagen, and telogen phases. These mesenchymal-epithelial interactions are assumed to stimulate germinative cells to proliferate, induce the anagen phase in the hair cycle followed by activation of stem cells, and consequently promote downgrowth of the hair follicles.

Stem cells were advocated by Cotsarelis et al.¹⁶ to be present in the bulge area of the infundibular region of the outer root sheath. The outer root sheath thus appears to play a major role in mature hair follicle formation. For hair fiber elongation following induction of the anagen phase in the hair cycle, activation of specific areas of the hair follicle, such as the bulge area in the outer root sheath and hair matrix in the hair bulb, is essential.

Table 4. Effects on hair growth

Group		The number of hairs			Hair diameter	
		Increase of hairs in 0.25 cm ²			Increased ratio of hairs (%)	
		Total hairs	Non-vellus hairs (>40 μ m)	Terminal hairs (>60 μ m)	Non-vellus hairs (>40 μ m)	Terminal hairs (>60 μ m)
Placebo	Mean	0.08	-1.63	-0.82	-4.07	-3.41
	SD	4.55	7.43	3.40	18.07	11.33
PB2*	Mean	6.68	5.79	1.99	6.52	3.06
	SD	5.53	4.85	2.58	11.89	8.13
	<i>p</i> **	<0.005	<0.005	<0.02	<0.1	<0.1

* PB2 = procyanidin B-2

** two-sample *t*-test

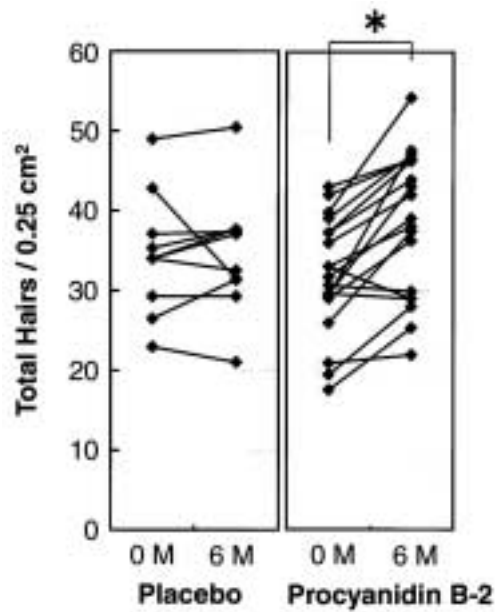


Figure 6. Change in hair density. The number of total hairs in the designated scalp area (0.5 cm square = 0.25 cm² area) after 6 months (6 M) of procyanidin B-2 treatment significantly increased over the baseline (0 M) figure for each subject ($*p < 0.001$, paired *t*-test); on the other hand, no significant difference was observed in the placebo controls (paired *t*-test)

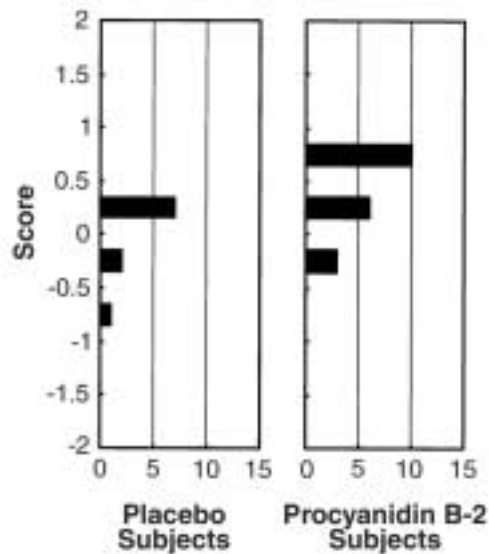


Figure 7. Evaluation by global photographic assessment. The score shows the average value of top side and back side. The scores of the procyanidin B-2 group were statistically higher than those of the placebo group (Mann-Whitney U-test, $*p < 0.05$)