华熙生物
BLOOMAGE BIOTECH

Bioyouth™-EGT Pro

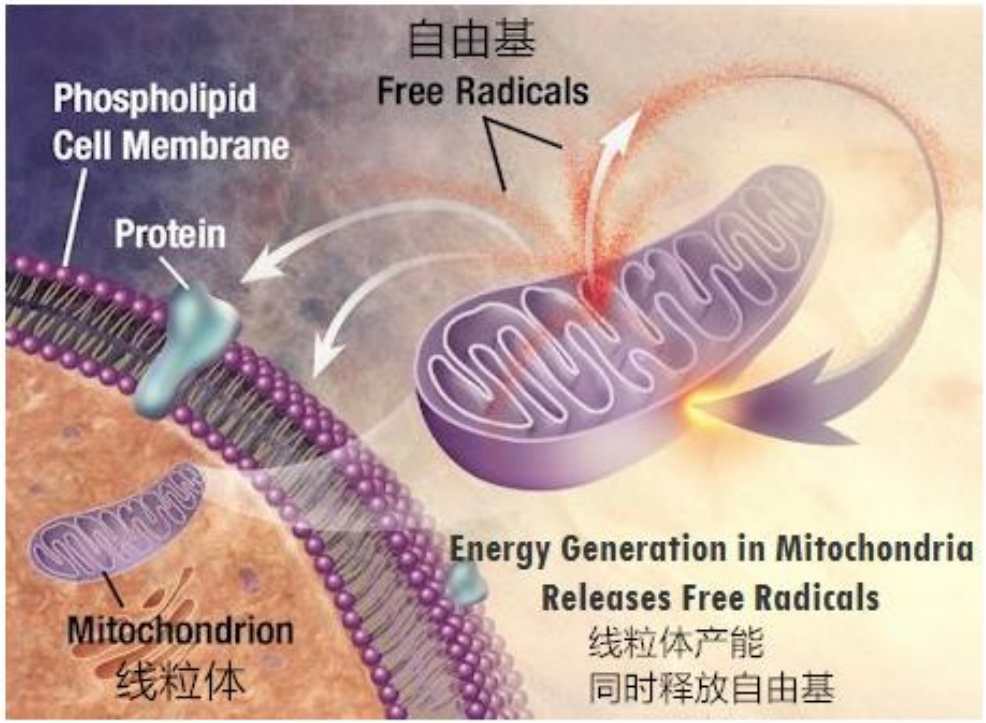
Super Active Ergothioneine

COSMOS

Antioxidant
Protect DNA and mitochondria
Anti-aging

Award：PCHI Fountain& Ringier
Patent：CN2019101570749
Bloomage Biotechnology Corp., Ltd.

Skin Aging and Mitochondria

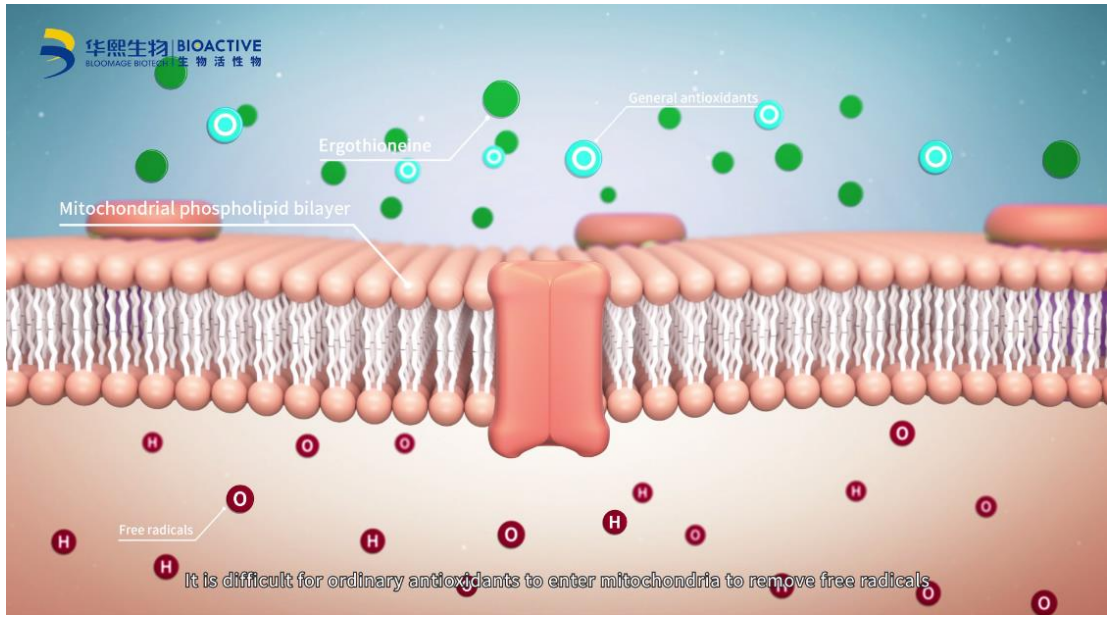


Mitochondria, as the main places for aerobic respiration and energy supply, generate a large amount of free radicals while outputting the energy. Excessive free radicals can cause mitochondrial apoptosis, leading to cell death and skin aging.

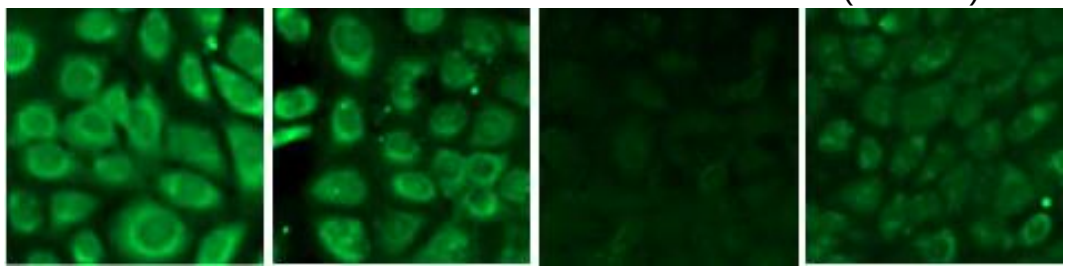
Recent studies suggest that the phenotypic features of photo-aged skin are the result of mitochondrial dysfunction and ECM degradation.

Ergothioneine and mitochondria

Few natural antioxidants can penetrate into mitochondria, while ergothioneine(EGT) can enter the cells and mitochondria through the specific transporter OCTN-1, directly scavenging ROS, and then provide efficacies of antioxidant and mitochondrial protection.



Control EGT (500nM) UVA(15J/cm^2) EGT(500nM)+ UVA(15J/cm^2)



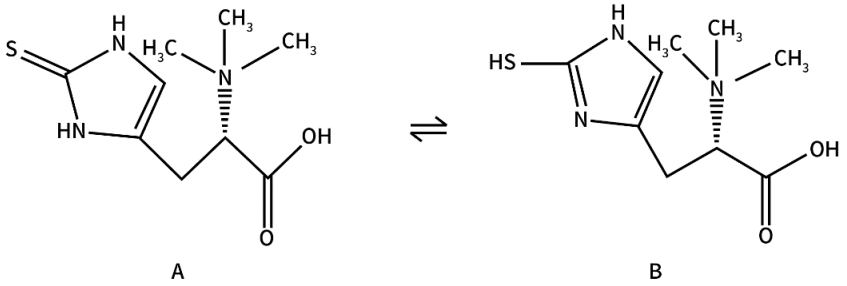
Method：Mito-tracker, HaCaT cells

Mito-tracker Green accumulates and displays green fluorescence in active mitochondria. The fluorescence intensity of UVA group was significantly lower than that of EGT+UVA, indicating that EGT can protect mitochondria from UVA damage [2].

Ergothioneine

In 1909, L-ergothioneine(EGT) was firstly isolated by Charles Tanret during his study of the ergot fungus, and soon EGT was also found in mushrooms and cyanobacteria. The human body cannot synthesize EGT, which can only be obtained through diet.

EGT has two structural tautomers of thiol and thione in the dissolved state. At physiological pH, EGT mainly exists in the form of **thione** in aqueous solution , so EGT is a very stable antioxidant compared with other naturally occurring thiols (for example, glutathione, N-acetylcysteine), and is less likely to spontaneously oxidize at physiological pH.



Two tautomers of EGT: A thione B thiol

Formula: $C_9H_{15}N_3O_2S$

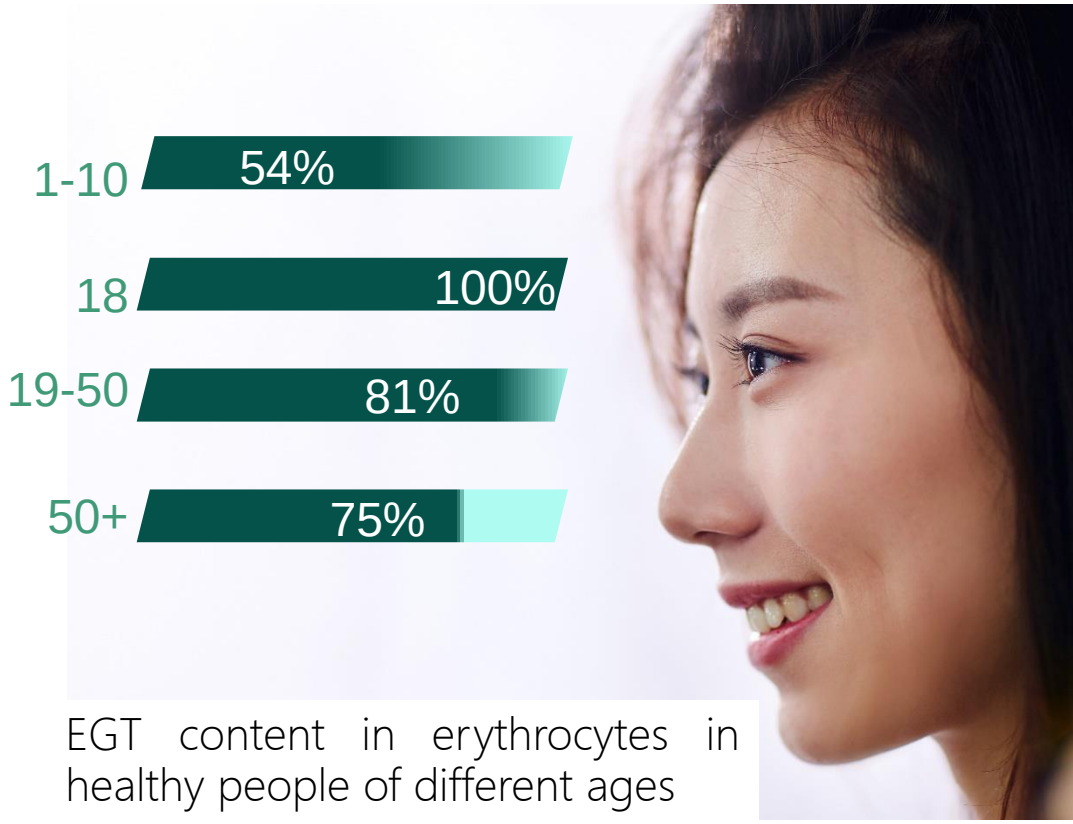
Molar mass: 229.298 g/ mol

CAS: 497-30-3

Ergothioneine in the human body

Closer examination of the distribution of EGT in the body reveals that the compound is preferentially accumulated in organs, cells and secretions predisposed to high levels of oxidative stress and inflammation such as liver, kidneys, erythrocytes, eye lens and seminal fluid[3]. After 18 years old, the content of EGT in human body decreases with age.

In July 2017, European Commission authorized the use of L-ergothioneine as a novel food ingredient.



EGT content in erythrocytes in healthy people of different ages

Academic Researches on Ergothioneine

Scavenge free radicals
Activate antioxidant
Inhibit super oxidant kinases
Anti-inflammatory
Chelate metal ions

Magnesium Ascorbyl Phosphate

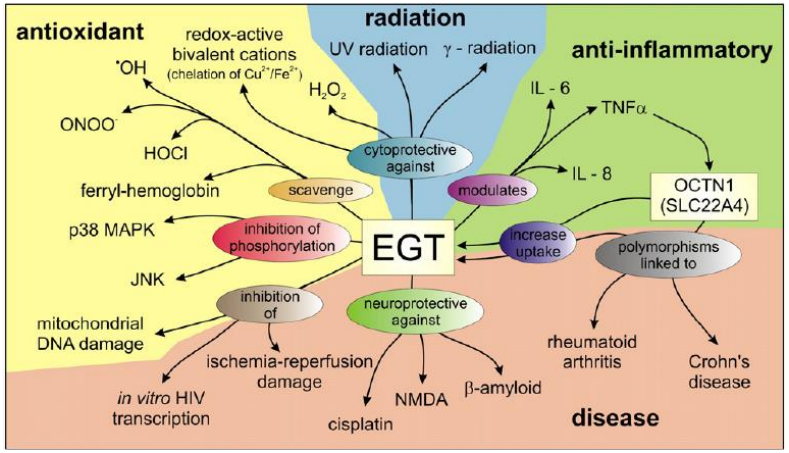
EGT is more efficient in inhibiting lipid peroxidation induced by alloxan than Magnesium Ascorbyl Phosphate [5].

Coenzyme Q10

EGT is more efficient in inhibiting lipid peroxide formation than coenzyme Q10[3].

GSH

EGT clears ROO- more than 550% better than GSH [4].



Idebenone

EGT has a greater ability to remove H_2O_2 and UVA-induced peroxides than idebenone[6].

TROLOX

The scavenging capacity of EGT towards hydroxyl radicals is 90% higher than the value obtained with trolox [4].

Bloomage Bioyouth™-EGT Series

Due to the existence of chiral carbon atoms, it is difficult to obtain L-Ergothioneine by chemical synthesis. Combining precise metabolic regulation and targeted control of multi-fermentation of *Hericium erinaceum* together with *Armillaria Matsutake*, Bloomage has successfully developed Bioyouth™-EGT series products.

Hericium erinaceum
It's a food and medicine, rich in protein, amino acids, vitamins and inorganic salts ,used to treat gastritis & gastric ulcer, and improve immunity, anti – aging.

Armillaria Matsutake
(*Tricholoma matsutake*)
It is called "the king of fungi". distributed in unpolluted high altitude virgin forest. They slowly grows on the fine roots of trees.

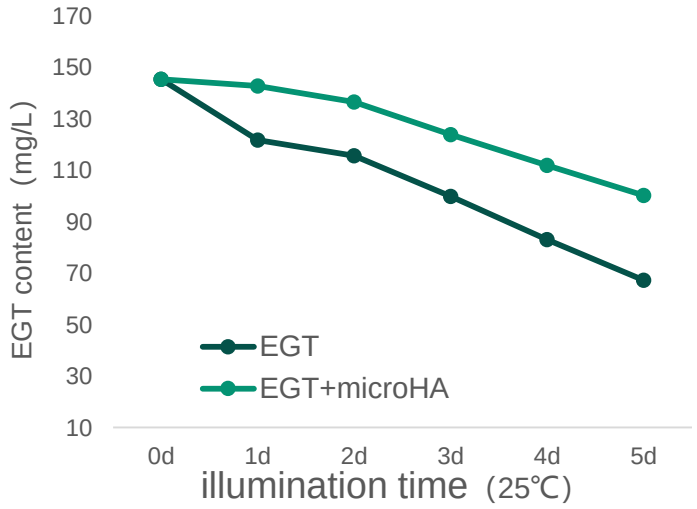
Bioyouth™-EGT

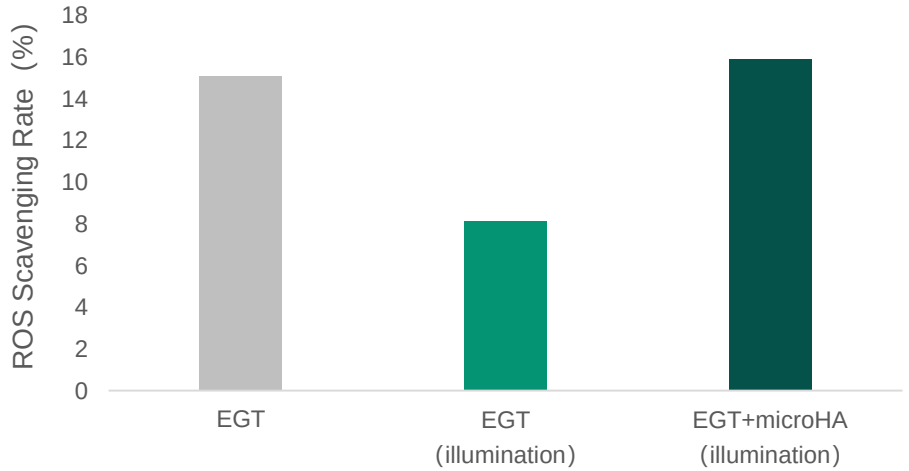
Bioyouth™-EGT has won the PCHI 2020 Fountain Awards and Ringier Technology Innovation Award 2020

Bloomage Bioyouth^{TM}-EGT Pro

The concentration of EGT has been increased by more than 10 times for Bioyouth^{TM}-EGT Pro, which is developed on the basis of Bioyouth^{TM}-EGT and spray dried together with microHA (M_W<5000Da) and trehalose in specific proportioning. Cytotoxicity test and skin patch test showed that Bioyouth^{TM}-EGT Pro can be used safely on human skin.

microHA can effectively protect the degradation of EGT against UV irradiation





microHA in Bioyouth™-EGT Pro

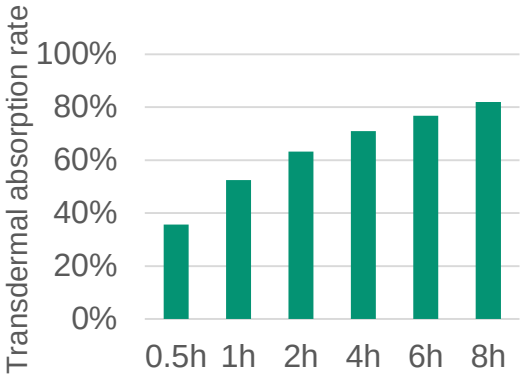
The 21st China Patent Gold Award：
ZL201210317032.5

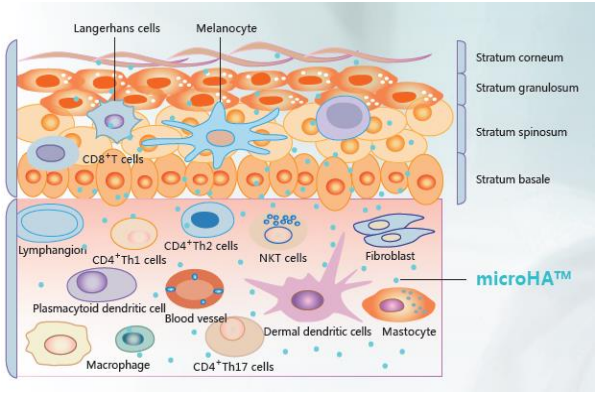
International patent
WO 2013/123791 A1

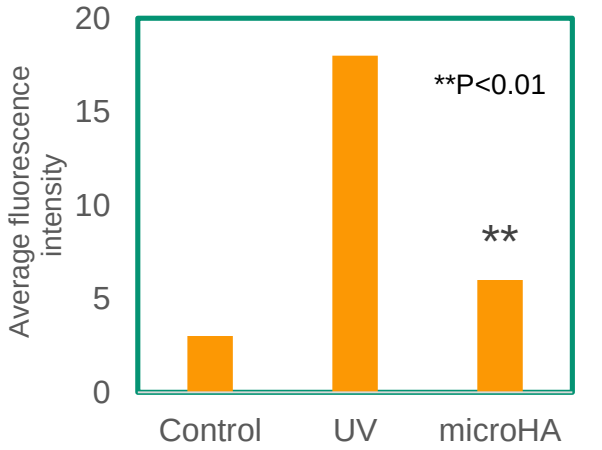
COSMOS certified

Extremely low molecular weight (≤5K Da)

Significant anti-inflammatory effect
Ultra-high repair activity







microHA（0.1%，m/m），test model:"UVA-L929"

microHA is easy to penetrate into the epidermis and dermis of the epidermis due to its extremely low molecular weight. The results show that, the absorptivity of microHA was 35.6% after 0.5 hour , 81.9% after 8 hours

Increase the Self-Defense of Skin Epithelium
Promote the function of the phagocytic cells and NK cells .
Form a coat of cells that acts as a barrier against cytotoxic lysis
Reduce the inflammatory reaction[7-9].

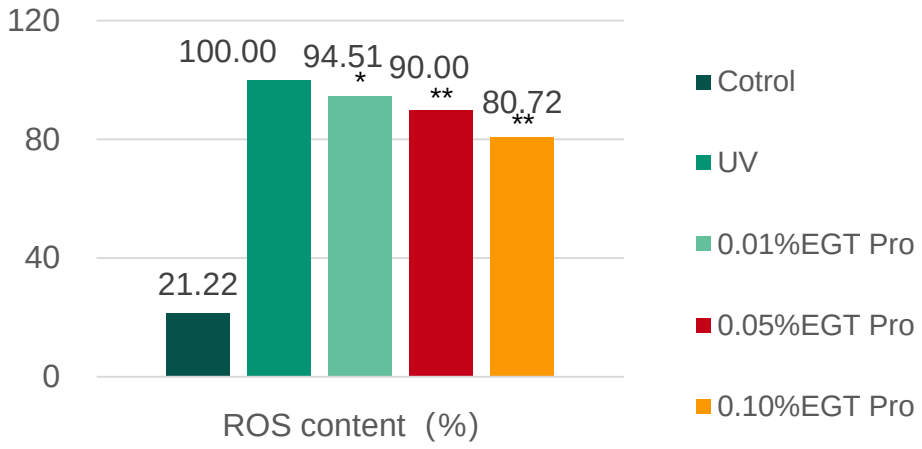
microHA can effectively remove the UV-induced ROS and reduce the inflammatory response. 1% microHA could reduce ROS by 81%.

Antioxidant Property-Scavenging ROS

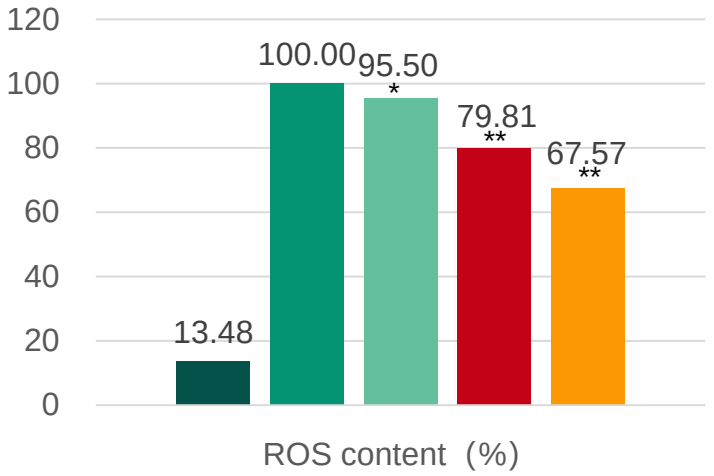
✓ Scavenge ROS induced by UV

HaCaT
*, $p < 0.05$; **, $p < 0.01$.

◆ Scavenge ROS- Protection



◆ Scavenge ROS- Repair



UV induced the production of ROS free radicals. Test shows that adding 0.1% EGT Pro before UV irradiation can reduce ROS production by **19%** , after UV irradiation can reduce ROS production by **32%** .

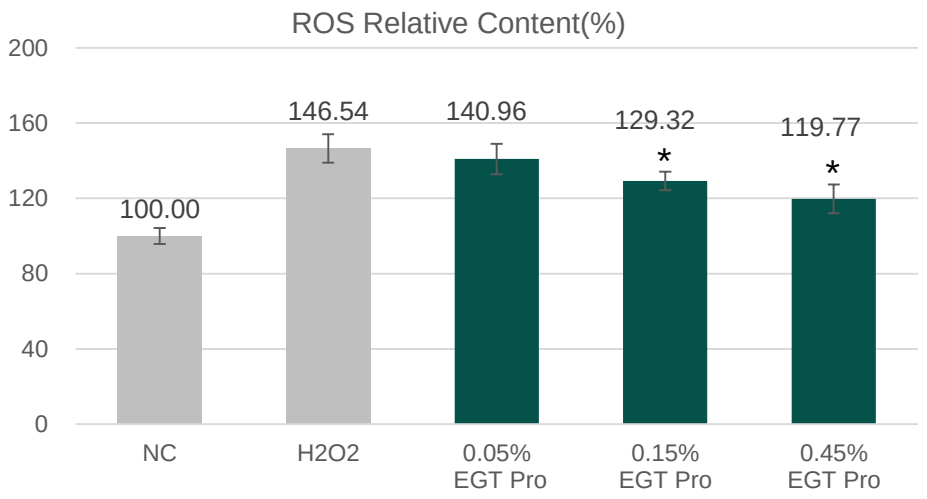
Antioxidant Property-Scavenging ROS

✓ Scavenge ROS induced by H_2O_2

Hydrogen peroxide(H_2O_2) is a major factor implicated in the free-radical theory of aging, based on how readily H_2O_2 can decompose into a hydroxyl radical and how superoxide radical byproducts of cellular metabolism can react with ambient water to form H_2O_2. These hydroxyl radicals in turn readily react with and damage vital cellular components, such as proteins, membrane lipids and DNA especially vital components of the mitochondria.

◆ Scavenge ROS- H_2O_2

*, $p < 0.05$

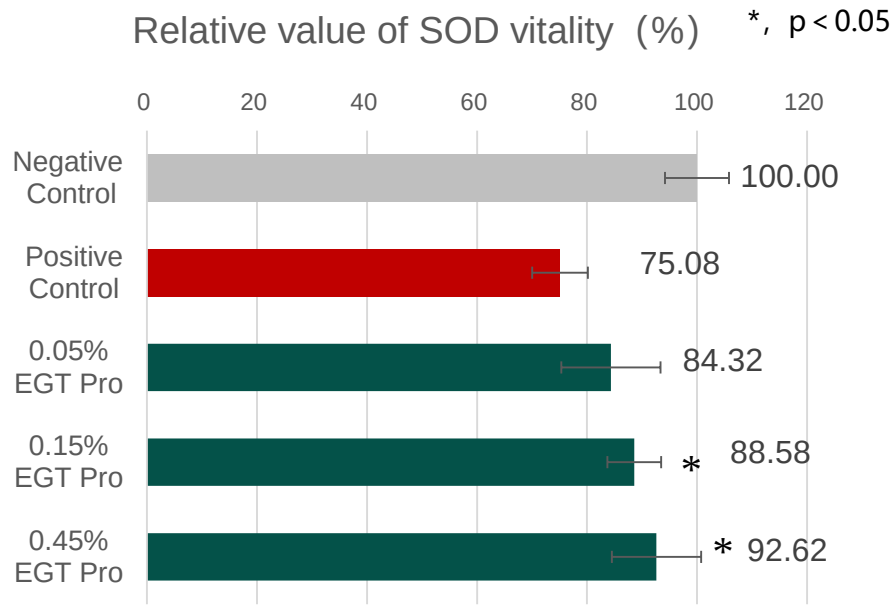


H_2O_2 induces ROS production in human fibroblasts. Test shows that 0.45% EGT Pro can scavenge ROS by **18%** compared with the positive control group.

Antioxidant Property-Increasing SOD Activity

$$O_2^- + O_2^- + 2H^+ \rightarrow H_2O_2 + O_2$$

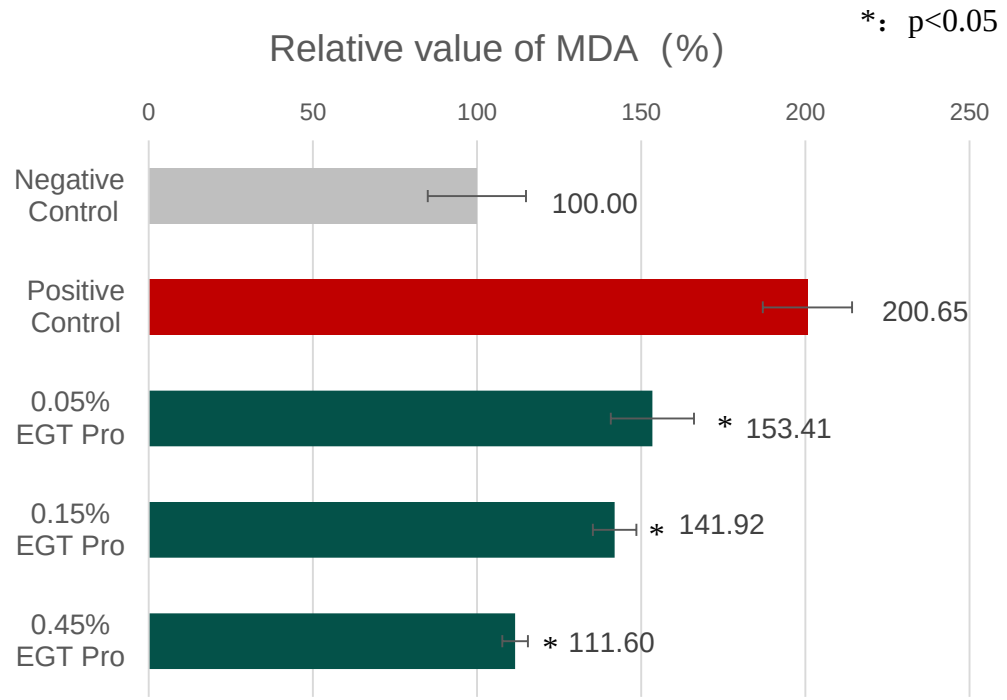
Superoxide dismutase (SOD) is an important superoxide anion free radical scavenging agent in organisms. It can specifically remove superoxide anion free radicals generated in the process of biological oxidation and contribute to delay aging. Therefore, SOD is considered as an important substance in the cell's antioxidant defense system. The immunogenicity, instability, inactivation and short half-life of SOD limit its application in clinic and skin care.



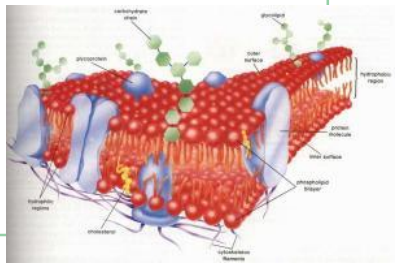
H_2O_2 inhibits the activity of SOD in human fibroblasts. Test shows that 0.45% EGT Pro can increase SOD activity by **23%** compared with the positive control group.

Antioxidant Property- Inhibiting Lipid Peroxidation

Lipid peroxidation is the oxidative degradation of lipids. It is the process in which free radicals “steal” electrons from the lipids in cell membranes, resulting in cell damage. Malondialdehyde (MDA) is a major by-product of LPO. It may have mutagenicity and carcinogenicity.. For instance, MDA reacts with deoxyadenosine and deoxyguanosine in DNA, forming DNA adducts to them.

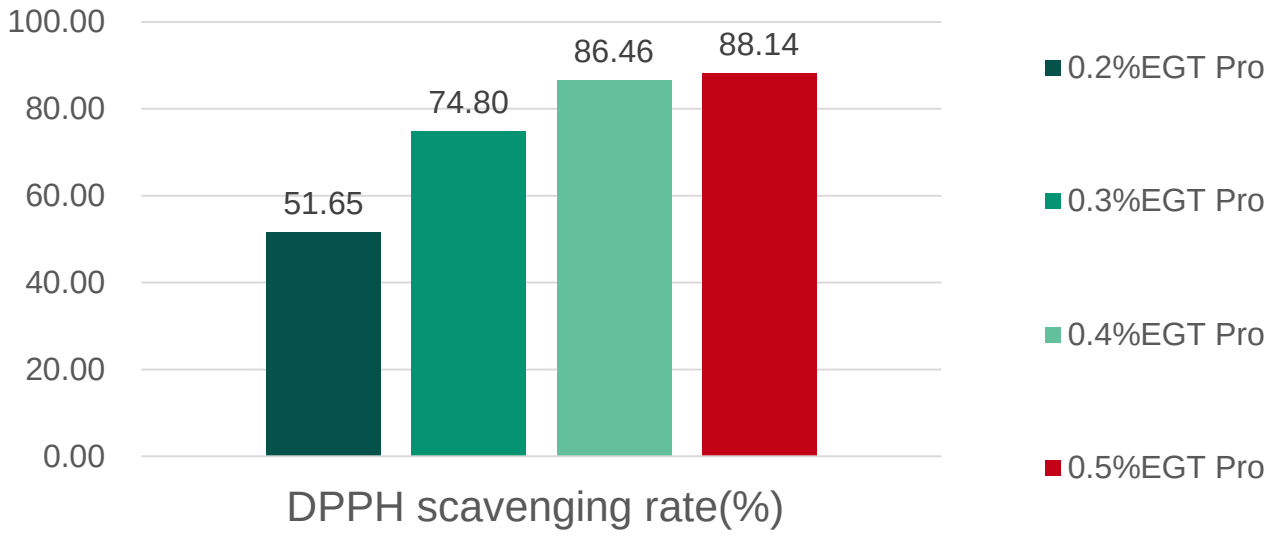


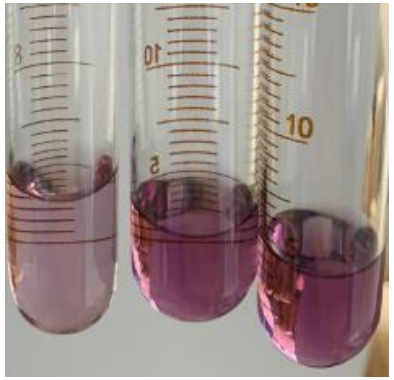
H_2O_2 stimulates cellular LPO and produces large amounts of MDA. Compared with the positive control group, 0.45% EGT Pro can reduce MDA content by **44%**.



Antioxidant Property-Scavenging DPPH

◆Scavenge DPPH free radicals





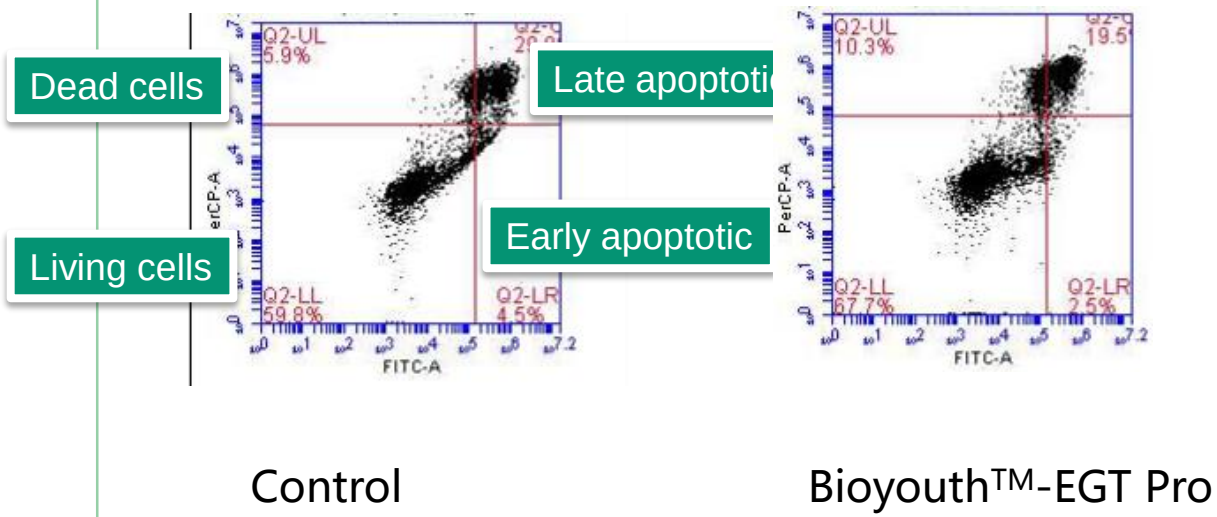
EGT Glutathione Control

Test shows that 0.5% Bioyouth™-EGT Pro can scavenge DPPH by 88%.

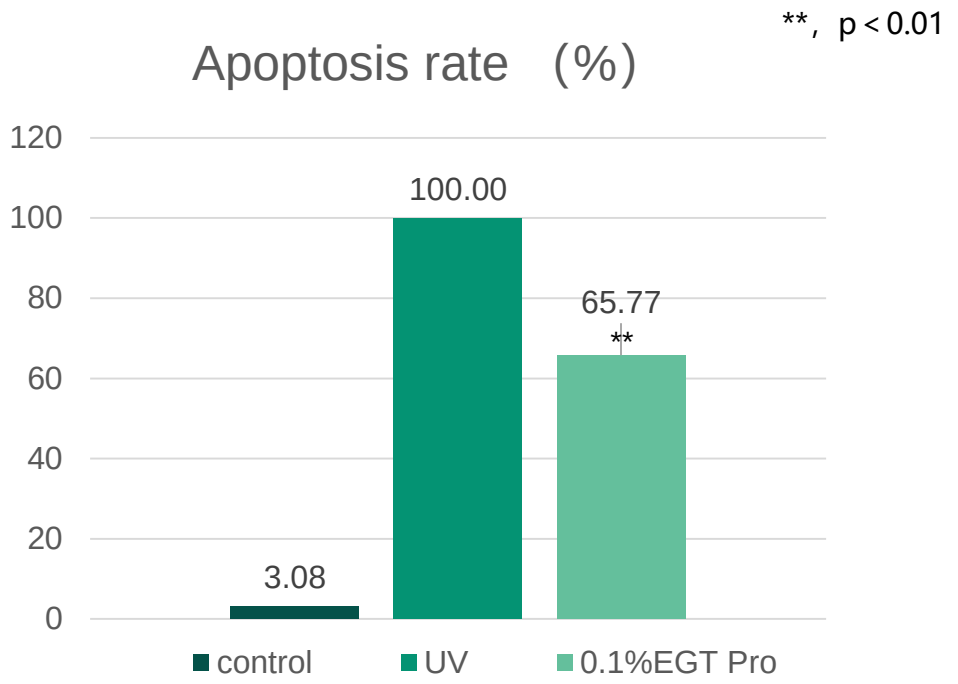
After heating, the DPPH scavenging rate of Bioyouth™-EGT was almost 7 times of glutathione at the same concentration.

Decreasing Cell Apoptosis

- ✓ Increase cell activity
- ✓ Decrease cell apoptosis



Control Bioyouth™-EGT Pro



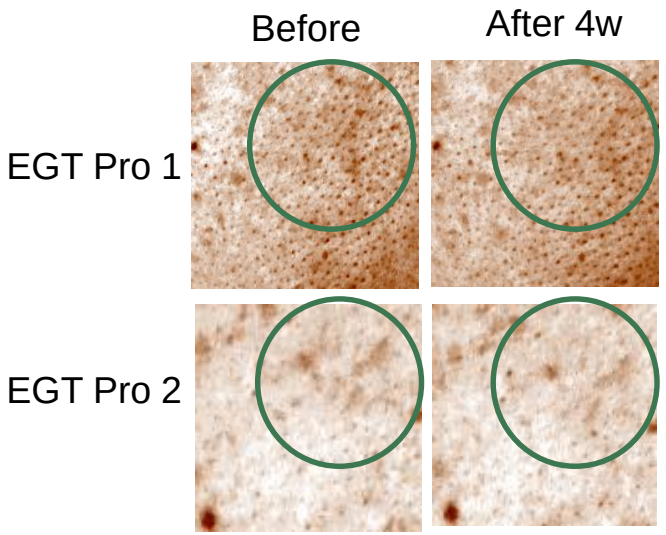
Cells tend to start its apoptosis because it senses cell stress or gets signals from other cells.
UVB irradiation increases HaCaT cell apoptosis. 0.1% Bioyouth™-EGT Pro can reduce the apoptosis rate by **34%**.

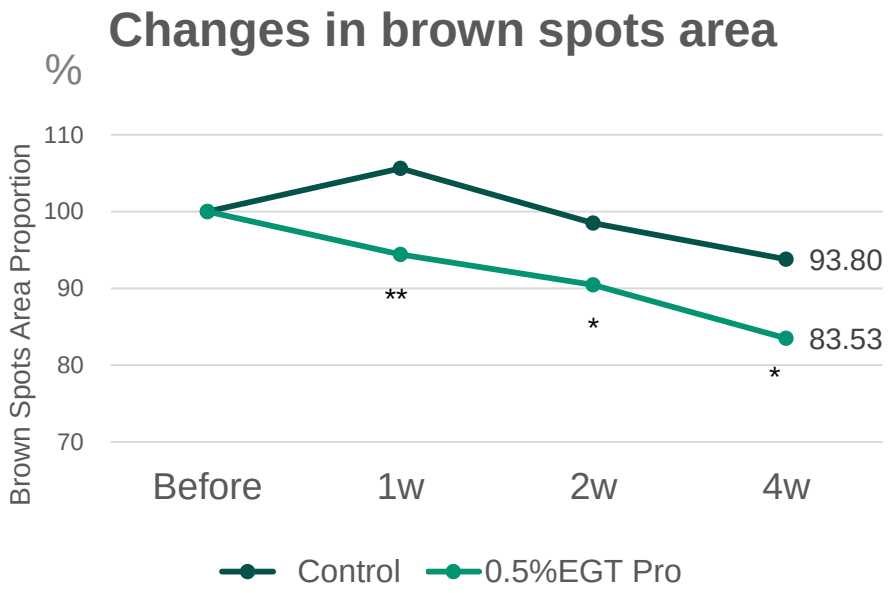
Flow cytometry : Single cells particles in suspension were quantitatively analyzed one by one at high speed by detecting labeled fluorescence signals.

Bioyouth™-EGT Pro *in-vivo* Test

✓Reduce Brown Spots

Samples：0.5% Bioyouth™–EGT Pro emulsion, half face, double blind





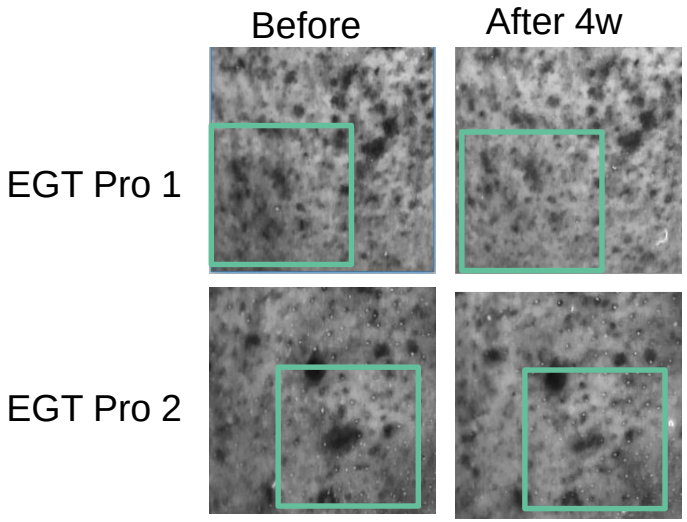
Compared to control group，EGT Pro can reduce brown spots by **10%** after 4 weeks application.

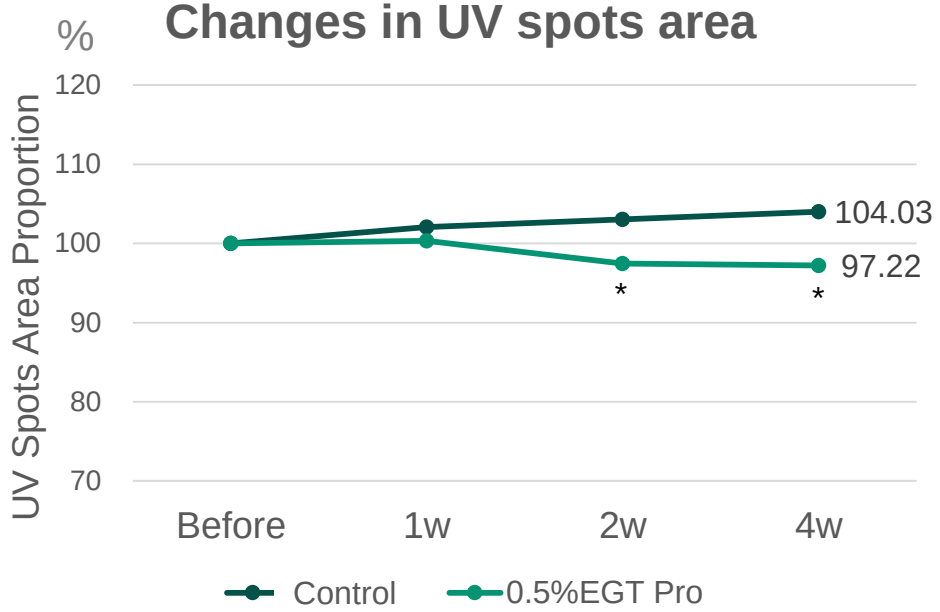
*，$p < 0.05$；**，$p < 0.01$。

Bioyouth™-EGT Pro *in-vivo* Test

✓Reduce UV Spots

Samples：0.5% Bioyouth™–EGT Pro emulsion, half face, double blind





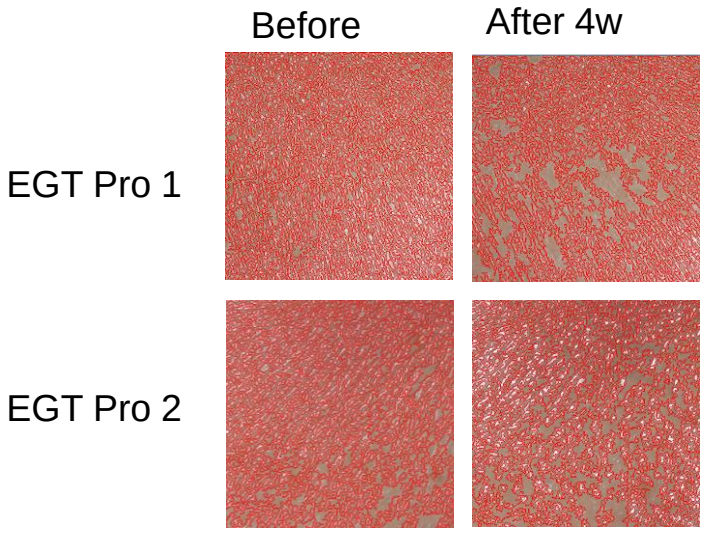
Compared to control group，EGT Pro can reduce UV spots by **7%** after 4 weeks application.

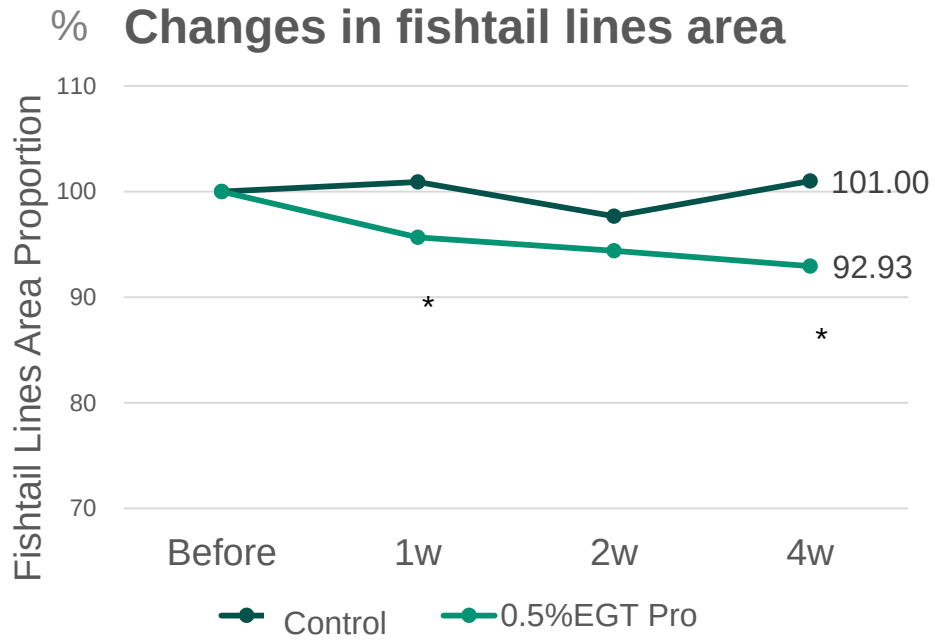
*，$p < 0.05$；**，$p < 0.01$。

Bioyouth™-EGT Pro *in-vivo* Test

✓Improve fishtail lines

Samples：0.5% Bioyouth™–EGT Pro emulsion, half face, double blind





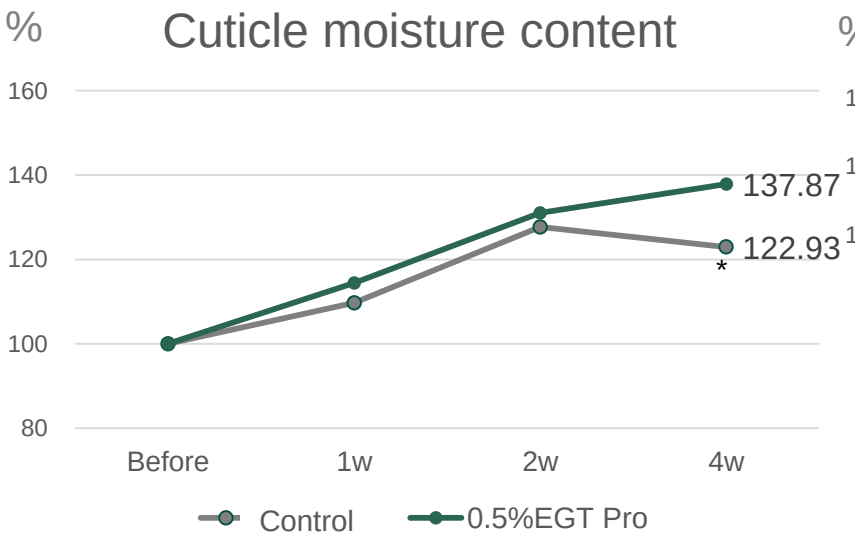
Compared to control group，EGT Pro can reduce fishtail lines by **8%** after 4 weeks application.

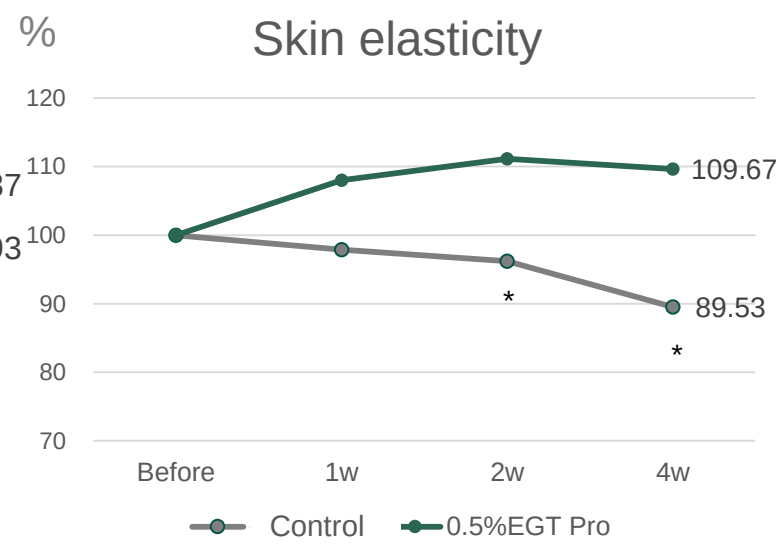
*，$p < 0.05$；**，$p < 0.01$。

Bioyouth™-EGT Pro *in-vivo* Test

✓Moisturize and improve elasticity

Samples：0.5% Bioyouth™ EGT Pro emulsion, half face, double blind





Compared to control group，EGT Pro can improve skin moisture by **15%,** improve skin elasticity by **20%.**

*，$p < 0.05$；**，$p < 0.01$。

Bioyouth™-EGT Pro Stability

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Temperature stability

0.1-0.5%
Heated at 40℃、60 ℃、80 ℃ separately for 1 hour

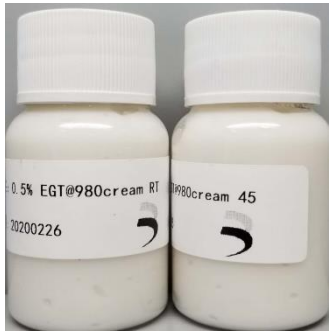
Solution color	No significant change
EGT content	
HA content	

pH stability

0.1-0.5%
Heat at 80℃ for 1 hour,
The solution is stable at pH 3-8,
Heat at 90℃ for 1 hour ,
The solution is stable at pH 4-7.

Application

Please use ion resistant thickening system and add Bioyouth™-EGT Pro at low temperature.



Before

After 4weeks

0.5% EGT Pro CARBOMER 980 lotion at room temperature (left) and 45℃ (right) for 4 weeks.(After emulsification)

Bioyouth™-EGT & Bioyouth™-EGT Pro

SKU	Bioyouth™-EGT	Bioyouth™- EGT Pro
INCI Name	Ergothioneine Armillaria Matsutake Extract Pentylene Glycol Ethylhexylglycerin	**Ergothioneine** **Armillaria Matsutake Extract** **Hydrolyzed Sodium Hyaluronate** **Trehalose**
Characters	Fermentation liquid	**Yellow or yellowish brown powder**
Recommended Dosage	0.5%-2%	0.1%-0.5%
Storage	Sealed, placed in the dark, cool dry place	Sealed, placed in the dark, cool dry place
Efficacy	Pan-himalayan source, environment friendly, Antioxidant, Anti-photoaging Protect DNA and mitochondria Improve skin laxity and dullness Smooth wrinkle	Pan-himalayan source, environment friendly, Antioxidant, Anti-photoaging Protect DNA and mitochondria Improve skin laxity and dullness Smooth wrinkle Better photodegradation resistance

Bioyouth™-EGT Pro Market Application

华熙生物 BLOOMAGE BIOTECH

EGT Products

Dior

Estee Lauder

Elizabeth Arden

Origins

DHC

SK-II

Givenchy

LA MER

PETER THOMAS ROTH

Paula's Choice

HA Products

QUOTE

[1]Bhupendra Singh1, Trenton R. Schoeb1, Prachi Bajpai1, Andrzej Slominski2,3 and Keshav K. Singh1,3,4. Reversing wrinkled skin and hair loss in mice by restoring mitochondrial function

[2] You-Cheng Hseu, Heng-Wei LO, Mallikarjuna Korivi, Yu-Cheng Tsai, Meng-Ju Tang, Hsin-Ling Yang, Dermato-protective properties of ergothioneine through induction of Nrf2/ARE-mediated antioxidant Genes in UVA-irradiated Human keratinocytes, Free Radical Biology and Medicine, 2015.05.026.

[3]Irwin K. Cheah, Barry Halliwell , Ergothioneine; antioxidant potential, physiological function and role in disease,Department of Biochemistry, Yong Loo Lin School of Medicine, National University of Singapore, 28 Medical Drive, Singapore.

[4] F. Franzonia, R. Colognatob, F. Galettaa, I. Laurenzac, M. Barsottid, R. Di Stefanod, R. Bocchettie,F. Regolie, A. Carpif, A. Balbarinid, L. Miglioreb, G. Santoro, An in vitro study on the free radical scavenging capacity of ergothioneine: comparison with reduced glutathione, uric acid and trolox, Biomedicine & Pharmacotherapy 60 (2006) 453–457

[5] KEI OBAYASHI, KOUJI KURIHARA, YURI OKANO, HITOSHI MASAKI, and DANIEL B. YAROSH, L-Ergothioneine scavenges superoxide and singlet oxygen and suppresses TNF- and IVIIVIP- expression in UV-irradiated human dermal fibroblasts, Cosmet. sci., 56, 17-27 (January/February 2005)

[6] Kelly K Dong, MS, Niusha Damaghi, BA, Jeannie Kibitel, MS, Matthew T Canning, MSc, Kenneth A Smiles, PhD, & Daniel B Yarosh, PhD, A comparison of the relative antioxidant potency of L-ergothioneine and idebenone, 2007 Blackwell Publishing •Journal of Cosmetic Dermatology, 6, 183–188

[7] Silvia Gariboldi, Marco Palazzo, etc.,《 Low Molecular Weight Hyaluronic Acid Increases the Self-Defense of Skin Epithelium by Induction of -Defensin 2 via TLR2 and TLR4》

[8]WANG Yu-kun , WANG Hai- jie , TAN Yu-zhenCd44 Mediates Effects Of Hyaluronan On Phagocytosis Of Macrophages》

[9]KE Chun - lin , LI Zuo - mei , ZENG Xiao - xiong , 《Immunomodulatory activity of low molecular weight hyaluronan on macrophages and T lymphocytes from mice》

Thank you谢谢

- Bloomage Biotechnology Corp., Ltd.