5. Solutions on Miraculin **

Summary

Rank	Solution	Solution proposed by	Non caloric or very low calorie	Stable at low pH (~pH 3.0)	Sensory profile closer to sugar	No licorice taste or Aftertaste	Thermal stability	Stable to UV exposure	Low cost	Robust supply chain	Solubility
16	Taste-modifying activity of miraculin	University of Tokyo; ERATO, JST; Suntory Institute for Bioorganic Research; Kyoto University; Imperial College London		Converts sourness to sweetness at acidic pH		Flat in taste under all the pH conditions employed					
27	Baïa Food's Miractin	**Baïa Food**		Activate in presence of acid		Uncertain aftertaste					
32	Mass Production of Miraculin in Tomato	University of Tsukuba, **Inplanta Innovations Inc**		pH of miraculin is ~4					Low cost	High yield in mass production	
34	Mass production of R-Miraculin in transgenic plants	University of Tsukuba, Iwate University						Miraculin concentrations were higher at low light intensity	Low cost	High yield in mass production	

Miraculin

Introduction

- **Miraculin is a glycoprotein found in red berries known as miracle fruit (Richadella dulcifica; synonym Synsepalum dulcificum),** produced by a tropical shrub native to West Africa.
- **Miraculin itself is not sweet, but it has a taste-modifying activity and is capable of converting sour taste to sweet taste. After chewing the red berry miracle fruit, lemons taste as sweet as oranges.**
- The name ‘miracle fruit’ was derived from this unique and attractive property, and the isolated active substance was named miraculin.
- **Miraculin accumulates only in the miracle fruit and its production begins 6 weeks after pollination, or at the turning stage, when the fruit’s color changes from green to orange,** the production peaks when the fruit is at the full-red stage.
- **Miraculin is a glycoprotein consisting of 191 amino acid residues and two sugar chains linked to Asn-42 and Asn-186**.
- The molecular weight of the single polypeptide chain calculated based on its amino acid sequence and carbohydrate content (13.9%) is 24.6 kDa.
- The nucleotide sequence encodes 220 amino acid residues, including a 29-amino acid signal sequence. This indicates that mature miraculin protein is processed by post-translational modification at its N terminus.
- Miraculin dimerization, mediated by a covalent linkage at Cys-138, is crucial for its taste-modifying activity at acidic Ph.
- **MCL is of particular interest because it has unique taste-modifying properties. Though flat in taste at neutral pH, it shows taste-modifying activity and converts sourness to sweetness at acidic pH.** Although this interesting sensory effect has been characterized, the molecular mechanism underlying the taste-modifying action of MCL is unknown.
- **Miraculin is able to elicit sweetness from various acids, such as HCl, oxalic acid, lactic acid, formic acid, acetic acid and citric acid; the sweetening effect is dependent on the sourness and the pH of the acid.**
- **The sweetening effect of a miraculin solution reaches its maximum level after being held in the mouth for approximately 3 min.**
- A concentration greater than 4 X 10-7 M is required for maximum effect, and the sweetness corresponds to 0.4 M sucrose solution.
- The taste-modifying effect of miraculin can be sustained for more than 1 h, although it depends on the concentration of the miraculin solution.

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5.1 Taste-modifying activity of miraculin **

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Taste-modifying activity of miraculin (1/2)

Solution

The solution to the problem is invented by University of Tokyo; ERATO, JST; Suntory Institute for Bioorganic Research; Kyoto University; Imperial College London

Non-patent Published in 2010

Application in food & beverages.

- **Miraculin (MCL) is a naturally occurring protein in the fruit of the West-African plant Richadella dulcifica.**
- It is a homodimer that consists of two glycosylated 191-amino acid polypeptides crosslinked by disulfide bonds.
- **MCL is of particular interest because it has unique taste-modifying properties. Though flat in taste at neutral pH, it shows taste-modifying activity and converts sourness to sweetness at acidic pH.** Although this interesting sensory effect has been characterized, the molecular mechanism underlying the taste-modifying action of MCL is unknown.
- Although sweet proteins exhibit no structural similarity, the importance of charged Arg and Lys residues was confirmed by the mutagenetic studies.
- MCL is a basic protein similar to other sweet proteins, but the role of the charged residues is not fully elucidated.
- In the case of another taste-modifying protein, neoculin, its sweetness depends on a structural change due to the pH change, although the histidine residues may always playing a key role.
- **Mutagenetic study of MCL using the Aspergillus oryzae expression system showed that the histidine residue His30, located at the interface of the two monomeric MCL subunits, is one of the candidate active residues.** As well as A. oryzae, other recombinant MCL (rMCL) expression systems have been reported previously.
- **To accelerate analysis of the structure–function relationships of the taste-modifying activity, however, it would be necessary to have a high-throughput expression system. The yeast Saccharomyces cerevisiae expression system has advantages as a high-throughput analysis system, and reported an advanced method useful for expressing proteins bearing various mutations.**
- **Using this advanced method, mutated proteins can be expressed within five days of mutation design. In addition, yeast can secrete folded proteins like A. oryzae.**
- These characteristics of yeast expression systems render them suitable for high-throughput mutagenetic analysis.
- **S. cerevisiae, however, has the disadvantage of relatively low expression levels of recombinant protein compared to other hosts. To alleviate this weakness, in this study we focused on codon usage and signal-sequence as the first step of the system.** These factors play an important role in high-level expression of secretory protein. Finally, through optimization of these factors, a 2 mg/l yield of rMCL in media was successfully obtained.
- **As the second step, the yeast rMCL was purified and its taste-modifying activity was analyzed. Although taste-modifying activity was not initially detected in the purified rMCL, it was significantly recovered by deglycosylation treatment. The structural effects of glycosylation on the taste-modifying activity are discussed, along with the results of the mutagenetic study.**

Materials and Methods

Step 1:
Codon optimization

Step 2:
Yeast transformation

Step 3:
Liquid expression

Step 4:
On-plate detection of secreted MCL

Step 5:
Purification and deglycosylation

Step 6:
Sensory analysis of the taste-modifying activity

Step 7:
Mutation analysis at the glycosylation sites

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Taste-modifying activity of miraculin (2/2)

Application

- ❑ Mutagenetic studies can broaden the scope of recombinant protein expression systems in very less time.
- ❑ The characteristics of different expression systems can made them suitable for high-throughput mutagenetic analysis.

Conventional Solutions

The molecular mechanism underlying the taste-modifying action of MCL was unknown.

The Saccharomyces cerevisiae expression system has advantages as a high-throughput analysis system, but compared to other hosts it is characterized by a relatively low level of recombinant protein expression.

Advantages

The yeast rMCL was purified and its taste-modifying activity was analyzed. Although taste-modifying activity was not initially detected in the purified rMCL, it was significantly recovered by deglycosylation treatment.

To alleviate this weakness, in this study optimization of the codon usage and signal sequence as the first step has been done.

Comment

- ➢ **A yeast expression system for MCL was constructed to accelerate analysis of its structure– function relationships.**
- ➢ **2 mg/l yield of rMCL was successfully obtained.**
- ➢ **Sensory taste evaluation showed that rMCL was flat in taste under all the pH conditions employed, taste-modifying activity similar to that of native MCL was recovered after deglycosylation.**
- ➢ Mutagenetic analysis revealed that the N-glycan attached to Asn42 was bulky in rMCL.
- ➢ The high-mannose-type N-glycan attached in yeast blocks the taste-modifying activity of rMCL.
- ➢ The bulky N-glycan attached to Asn42 may cause steric hindrance in the interaction between active residues and the sweet taste receptor hT1R2/hT1R3

Source

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5.2 Baïa Food’s Miractin **

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Solution

Baïa Food's Miractin (1/4)

The solution to the problem is invented by Baïa Food

Application in food & beverages.

- Medicinal Gardens SL, known commercially as Baïa Food, is an Innovative SME with a strong R&D investment. **Baïa is a Spanish foodtech known to the public since 2017. The company was founded in 2013 by Guillermo Milans del Bosch and Loan Bensadon.** The two Co-founders began their economic activity in 2015.
- **Baïa was created with the initial purpose of being an accessory line to that of the miraculin that would allow billing to be able to be a beneficiary of public aid and to serve as a reference against the food industry, thus serving a second purpose, the of networking, since the idea was "to be able to present the project of the miraculin", and to know the demands of the consumer.**
- Direct from Ghana (Africa), The miracle berry is able to hide sour and bitter flavors and stimulates sweets in food and beverages. It seems to be too good to be true that it has cost this startup eight years, another business and more than 750,000 euros invested in R&D. **This natural sweetener "binds to the sweet taste receptors that we have on the tongue and activates them in the presence of acid, making any food and drink with a pH lower than 6 perceived as sweet", therefore its potential to reduce the consumption of sugar and artificial sweeteners is infinite.** The founders thought that the miraculin project was not going to take more than three years. However, passing the Novel Food regulation as it is categorized as such by the EU according to Regulation (EU) 2015/2283 has not been easy. Guillermo, hope this year to obtain the authorization of the miraculin and achieve the first sales of this line of business, whose commercialization will be carried out at the beginning under the category of food supplement. **The only objective of the startup is to offer a natural and healthy alternative to artificial sweeteners and sugar.**
- Along the way, the co-founders have managed to build an e-commerce that grew from 2019 to 2020 of more than 300%, has forged an ethical and sustainable supply chain at source, and they have carried out endless tests to demonstrate the safety of food. The Baïa Food team discovered the Synsepalum dulcificum plant in Ghana back in 2013 and identified the "untapped potential" of its antioxidant-packed fruits, dubbed miracle berries, as a natural sugar-reduction strategy. **Recently, the European Food Safety Authority (EFSA) has published a scientific opinion concluding that the dried miracle berry (DMB) is safe for use in food supplements at the maximum intake level of 0.7 g per day for adults, excluding pregnant and lactating women.** The company has a DTC business line with a market place and offers organic superfoods, proprietary plant-based powder blends and is developing with an industry partner ready-to-drink organic plant-based protein beverages. **The company is presently in the process of authorization of Miractin®, a proprietary standardized extract (>1.5% miraculin) which has proved the safety in a battery of 5 in vitro (mutagenicity & micronucleus) and in vivo studies (micronucleus, acute and sub-chronic toxicity) and the Novel Food dossier was submitted to the European Food Safety Authority at the end of 2018. The sweetness with the miraculin is achieved in two steps: the dehydrated berry is consumed first, for example in the form of candies, and then the food whose acid flavor you want to transform to sweet, such as kefir or a yoghurt. The effect is immediate and can last between 30 minutes or an hour and the maximum peak in the ability to modify the flavor is reached within a minute and a half.**

Source1 Source2

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5.3 Mass Production of Miraculin in Tomato **

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Mass Production of Miraculin in Tomato (1/4)

Solution

The solution to the problem is invented by University of Tsukuba, Inplanta Innovations Inc

Non-patent Published in 2010

- **The reference here discloses about breeding suitable tomatoes for closed cultivation system by crossing the 56B and "Micro-Tom" varieties and producing miraculin in these crossed lines.**

Application in food & beverages.

Plant Material:

- **The transgenic tomato line 56B (upright type, "Moneymaker") possesses the miraculin gene driven by the CaMV 35S promoter,** and the recombinant miraculin protein accumulates in all plant tissues. **The homozygous line from the T3 generation of 56B was crossed with a dwarf tomato cultivar ("Micro-Tom")**, and the transgenic plants were grown in a netted greenhouse and allowed to self-pollinate. **Plant size, miraculin accumulation, and self-pruning were used as selection indicators for F2 plants.** The two selected lines were named cross no. 1 and cross no. 2, and they were bred to the F7 and F6 generations, respectively, by self-pollination. Fruit yield was used as a selection indicator in subsequent generations. The plants of 56B and the crossed lines were cultivated in a closed cultivation system.

Construction of the Tomato Cultivation System:

- **Transgenic tomato seeds of 56B (T7), cross no. 1 (F7), and cross no. 2 (F6) were germinated on Petri dishes covered with moist filter paper at 25/20 C (light/dark), with 16 h of light from a fluorescent lamp at 450 µmol/m2 /s (photosynthetic photon flux) and 8 h of dark in the plant factory. The CO2 concentration was maintained at 600 ppm.** One week after sowing, seedlings were transplanted to rockwool cubes (5 cm 5 cm 5 cm) and grown in a Naeterrace seedling raising system. Each day, **the plants were provided with a nutrient solution containing 565 mg/L NO3 -, 15.7 mg/L NH4 þ, 202.2 mg/L PO3 -, 218.4 mg/L Kþ, 19.9 mg/L Mg2þ, 95.0 mg/L Ca2þ, and micronutrients.** Each seedling was then transferred to a two-layer cultivation system. For 32 days after germination, the plants were grown under the same conditions as those used in the seedling raising system, except for the nutrient solution. **Otsuka-A nutrient solution, with an adjusted electrical conductivity of 1.8 dS/m, was supplied every day in the two-layer cultivation system. The planting densities of 56B and the crossed lines were 13.3 and 26.7 plants/m2 , respectively**. The 56B plants were pruned, leaving three leaves above the first truss, and axillary buds were removed during cultivation. Axillary buds of cross no. 1 plants were also removed approximately two weeks after transplanting. Extra leaves were removed from 56B and cross no. 1 plants. There was no need to defoliate cross no. 2 leaves.

Source

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Mass Production of Miraculin in Tomato (2/4)

Tests

Analysis of Tomato Fruit Quality and Yield:

- **Fruits were harvested once per week from day 48 to day 104 after transplantation to the two layer system.** Fruits with blossom-end rot were removed, and dehiscent fruits were counted. **The weight, length, and diameter of each fruit were measured.** The fruit yield per area per year (kg FW/m2 /year) was calculated based on the fruit yield per plant (g FW/plant), planting density (plants/ m2), and growing period per year (days/year) in the two-layer system. **Separation of Fruit Tissue** - To measure the fresh weight and detect the miraculin protein in different tissues of the tomato, **the fruits were separated into two parts: the pericarp and all other tissues.** The other tissues were first separated from the fruit, which was cut into halves. The pH was measured in part of the pericarp. **The pericarp was separated into the exocarp and mesocarp by removing the exocarp after the fruit was dipped into liquid nitrogen for a few seconds.** The fresh weight of each part was measured, and the separated fruit tissues were immediately frozen with liquid nitrogen.

Immunoblot Analysis, Enzyme-Linked Immunosorbent Assay (ELISA), and Protein Assay:

- **The accumulation and concentration of the miraculin protein in 56B and the crossed lines were determined using immunoblot analysis and ELISA. Collected fruit tissues described above were ground to powder in liquid nitrogen. The powder (0.1 g FW) was thawed in 200 µL extraction buffer consisting of 20 mM Tris-HCl (pH 8.0), 0.5 M NaCl, and 2% polyvinylpolypyrrolidone.** The extracts **were centrifuged at 15000 rpm for 20 min at 4 C**, and the supernatant was used for immunoblot and ELISA analyses. The extracts (the equivalent of 0.5 mg FW per lane) were separated by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and transferred onto Hybond-P polyvinylidene fluoride membranes. Miraculin accumulation was determined using immunoblot analyses. **To measure the concentration of miraculin, 100 µL of 500-, 1000-, or 2000-fold dilutions of supernatant were applied to a 96-well plate, and various concentrations of purified miraculin protein were used as standards.** ELISA was conducted. The soluble protein concentrations in the fruit tissues were determined using the BCA Protein Assay Kit.

Histochemical Analysis:

- **Fruits at the green stage were sliced at the appropriate size to observe the tissue in 56B and the crossed lines**. Transverse slices (100 µm) were prepared using a Leica vibratome. **The separated sections were stained with safranin and observed under an optical microscope.**

Source

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Mass Production of Miraculin in Tomato (3/4)

Results

Fruit Yields in 56B and the Crossed Lines:

- **The fruit yield per plant of cross no. 1 was higher than that of 56B, and the fruit yield of cross no. 2 was similar to that of 56B.** The fruit yields per plant of 56B and cross no. 1 were greatly reduced 76 days after transplanting into the two-layer cultivation system because their axillary buds were removed. **The fruit yield per plant of cross no. 2 increased linearly until the end of the cultivation period. The maximum fruit yields per area per year for 56B, cross no. 1 and cross no. 2 were 26.2, 73.6, and 45.9 kg FW/m2 /year,** respectively. The maximum fruit yield of cross no. 1 per area per year was much higher than that of 56B, and that of cross no. 2 was also higher than that of 56B. **Thus, cross no. 1 was the best line with respect to fruit yield.**

Miraculin Accumulation and Concentration and pH level in the Pericarp of Fruits of 56B and the Crossed Lines:

- **The miraculin concentrations in the pericarp of the crossed lines were also approximately 2.5 times higher than that of 56B.** These results show that the crossed lines performed better with respect to miraculin production in the same amount of pericarp than 56B. The native miraculin protein was stable under acidic conditions, so the pH levels in the transgenic tomato fruits were measured. **The pH levels of 56B and the crossed lines were 4.15 and approximately 4.05, respectively.**

Miraculin Accumulation and Concentrations in Various Fruit Tissues of 56B and the Crossed Lines:

- Fruits were divided into three parts: the exocarp, the mesocarp, and all other tissues. **In the fruits of all lines, the miraculin accumulation and concentration in the exocarp were much higher than in the other tissues. In the crossed lines, miraculin accumulation levels and concentrations in the mesocarp and other tissues were higher than those in the 56B line.**

Ratios of Fresh Weight of Each Tissue, Soluble Protein Concentration, and Histochemical Analysis in 56B and the Crossed Lines:

- **The weight percentages were determined from the average weight of eight different fruits. The exocarp weight percentages in 56B, cross no. 1, and cross no. 2 were 3.2%, 4.3%, and 6.1%, respectively. The weight percentages of the mesocarp and other tissues were similar among all lines.** The soluble protein concentration in the exocarp of all lines was higher than in the other tissues. The soluble protein concentration in the mesocarp of the crossed lines was similar to that in 56B, and the soluble protein concentration in the other tissues of the crossed lines was higher than that in 56B. The histological data showed that the exocarp cells were smaller than the mesocarp cells in 56B and the crossed lines. **The mesocarp cells of 56B were larger than those of the crossed lines.**

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Mass Production of Miraculin in Tomato (4/4)

Application

- ❑ The method is applicable for mass production of miraculin in a plant factory.
- ❑ The method is also cost effective and produces better yield of miraculin.

Conventional Solutions

The conventional production method has less yielding capacity

Advantages

The method is capable in mass production of miraculin in a plant factory with decreased production cost and increased miraculin production.

Comment

- ➢ The mass production of miraculin is possible by using this method in a plant factory.

Source

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5.4 Mass production of Miraculin in transgenic plants **

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Mass production of Miraculin in transgenic plants (1/3)

Solution

The solution to the problem is invented by University of Tsukuba and Iwate University

Non-patent Published in 2011

Application in food & beverages.

Hosts for the production of recombinant miraculin:

- **Recombinant miraculin produced in E. coli was found to have no taste-modifying activity** even though the protein was detected by SDS-PAGE and Western blot analysis.
- The production of recombinant miraculin was attempted again in E. coli by Matsuyama. **They succeeded in producing an active miraculin dimer. However, its activity was only one-sixth that of native miraculin. This result showed that the glycosylation of miraculin is crucial for its protein folding and/or stability.**
- **An active form of recombinant miraculin was also produced using A. oryzae (which has glycosylation capacity) as a host strain.**
- **In the Saccharomyces cerevisiae yeast system, recombinant miraculin was produced after optimizing codon usage and the signal sequence, although the activity was only detected when the sugar chains were removed.**
- The activity of recombinant miraculin in both A. oryzae and S. cerevisiae was evaluated at one-fifth the concentration of native miraculin.
- **These results suggest that not only is glycosylation important, but the types of sugar chains are also crucial for high and stable activity.**
- **Transgenic tomatoes were found to be the most suitable hosts for recombinant miraculin production from these plants.**
- **Tomato also has an advantage in the established Agrobacterium-mediated transformation protocol using cotyledon explants from seedlings, and the transformation efficiency exceeds 40% of the explants.**
- **The transformants are able to be obtained for 3 months after the seed germination.**

Improving recombinant miraculin accumulation in transgenic tomato fruit

- **Accumulation profile of recombinant miraculin in transgenic tomato fruit -** Recombinant miraculin accumulates to high levels in the overripe fruit during fruit development when the miraculin gene is driven by the 35S promoter. Among fruit tissues, miraculin levels in the exocarp, or epidermis were found to be extremely high at 928 lg/g FW compared to below 110 lg/g FW in other tissues. **The deduced amino acids of miraculin include an N-terminal signal sequence of 29 amino acids. Miraculin protein is transported to and accumulates in the intercellular layer space of both miracle fruit and transgenic tomatoes.** The amount of miraculin per dry weight, which is less affected by cell size, hardly differs between the exocarp and other tissues. **Therefore, the researchers speculate that the high miraculin accumulation in the exocarp is caused primarily by cell size which influences the amount of intercellular layer space.**

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Mass production of Miraculin in transgenic plants (2/3)

Solution

- **Cultivation in a closed system**
  - **Closed cultivation systems also have other advantages:** it is easy to prevent the spread of transgenic plants and pollen to the external environment, and at the same time, the plants are protected from disease and pests from the external environment. However, the limited space available for cultivation and the high operating costs become bottlenecks. **The miraculin content in the tomato fruit significantly increased to 343 lg/g FW of pericarp (including exocarp and mesocarp), which is approximately 2.5 times higher than that of 56B, due to the effect of genetic background modification. This effect was confirmed by the 1.9-fold higher weight ratio of the exocarp in cross no. 2 than in 56B, and no. 2's high miraculin concentration in the mesocarp relative to that of 56B.** The high operating costs of closed cultivation systems mainly accrue from the electric bills due to the artificial lighting and air conditioning. To reduce electricity costs, it is important to understand the effects of light intensity on both tomato fruit yield and the production of recombinant miraculin. **Miraculin concentrations were higher at low light intensity, but fruit yield was higher at strong light intensity. Consequently, the miraculin production per unit area was highest at PPF300, but the miraculin production per unit of energy was best at PPF100. Thus, these results indicate that it is necessary to select a suitable light condition on the basis of market demand and the sales price of recombinant miraculin.**

Table 3 Comparison of transgenic tomato yield and recombinant miraculin production under different cultivation condition in closed cultivation systems

Transgenic tomato	PPF (μmol/m²/s)	CO_2 concentration (ppm)	Finally planting density (plants/m²/layer)	Light period (h/day)	Days to harvest of all tomatoes (days after sowing)	Miraculin concentration (μg/g FW)	Number of layers (at same height)[a]	Fruit yield (kg FW/m²/year)	References
56B	450	600	14.1	16	109	81	2	35.5	Hirai et al. (2010a)
	600	600	14.1	12	105	82	2	44.1	
	600	600	14.1	16	104	87	2	46.3	
56B	450	600	13.3	16	90	140	2	26.2	Kato et al. (2010)
Cross no. 1	450	600	26.7	16	90	367	2	73.6	
Cross no. 2	450	600	26.7	16	104	343	2	45.9	
Cross no. 2	100	1,000	44.4	12	95	472	3	25.5	Kato et al. (2011)
	200	1,000	44.4	12	86	362	3	51.0	
	300	1,000	44.4	12	84	272	3	83.1	
	400	1,000	44.4	12	79	211	3	91.5	

PPF photosynthetic photon flux

[a] It means the number of installation available layers on the height of closed cultivation system

Source

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Mass production of Miraculin in transgenic plants (3/3)

Applications

- Miraculin finds novel uses as a component of nutritionally enhanced snacks and drinks with the possibility of widespread consumer acceptance and new marketing opportunities.
- Industrial uses of recombinant miraculin from transgenic tomatoes encompass the consumption of unprocessed raw fruit and processed fruit, use as a food additive and as a reagent for research and development.
- The taste-modifying function can potentially be used not only as a low-calorie sweetener but also as a new seasoning that could be the basis of a new dietary lifestyle.
- Future studies will assess the safety of recombinant miraculin: it is essential to investigate such aspects as its toxicity, allergenicity, digestibility, thermal stability, insertion position in the host genome and processing status.

Conventional Solutions

- *In conventional experiments, microbial and animal systems were used as host and therefore restricted the use of types of recombinant proteins.*

Advantages

- *The advantage of using an edible plant as a host lies in the fact that several processed and unprocessed types of recombinant proteins can be used and can minimize purification costs.*
- *Miraculin has a taste-modifying function that changes a sour taste into a sweet taste. This is helpful for preventing lifestyle diseases in that it can be used as a low-calorie sweetener or as an additive for foods targeting diabetics.*

Comment

- The characteristics of miraculin and recent advances in its production using transgenic plants are summarized, focusing on the suitability of plant species as expression hosts, the cultivation method for transgenic plants, the method of purifying miraculin and future advances required to achieve industrial use.
- Transgenic plants are presumably the most suitable hosts for recombinant miraculin protein production with respect to post-translational modifications such as glycosylation.
- The transgenic tomato has many advantages, including genetic stability, good accumulation levels and good fruit-bearing capacity, and it is readily available to procure as a raw food.
- Thermal stability of the recombinant miraculin protein is also crucial information for more optimization of industrial use.

Source

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