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1. 서론

- ✓ 실크 피브로인(SF;Silk Fibroin)의 화장품 및 건강 기능성식품 개발 및 응용^{1,2}
 - 4차 산업 기반 산업용 단백질 생산에 대한 관심이 높아지고 있는 추세
 - SF는 우수한 생체 적합성, 생분해성, 세포 부착 및 증식 효과 향상, 세포 사멸 방지 등의 효과가 알려져 있음.
 - 세리신 단백질은 피부 노화 방지, 보습 효과, 멜라닌 생성 효과가 알려져 미용재료, 화장품 재료, 음식 등에 다양한 활용 등이 보고 되고 있음.
 - SF의 산업적 이용:
 - 식품 소재 및 기능성: 아미노산의 건강 기능성, 건강 기능성 펩타이드, 실크 펩타이드의 아미노산 조성,
 - SF 분말의 건강 기능성: 항산화 작용, 간 기능 개선, 인슐린 분비 촉진 작용, 혈중콜레스테롤 상승 억제, 신경계 질환 예방 및 개선, 실크 단백질의 식품화
 - 피부 친화형 소재 이용: 기능성 화장품, 색소의 고정화, 고급 기능성 미용 비누, 두발 염모제
 - 생체 의용 소재 이용: 창상 피복제, 수술용 색전 물질, 수술용 봉합사, 완충형 약물 방출 제제
 - 산업용 소재 이용: 섬유 가공제, 부직포 류, 실크 플라스틱, 고흡수성 수지.

2. 연구 동향

✓ Novel Beneficial Functions of Silk Protein, Sericin for Health¹

- Silk protein의 한 종류인 sericin은 변비증상과 장염을 방지하고 무기물 생체이용률을 높이며 혈당을 낮게 해주는 효과를 보임.
- 1~4% sericin이 있는 음식을 동물들이 먹었을 때, 부작용을 보이지 않았으나, 6% sericin이 담긴 음식을 먹었을 때는 이 단백질의 보수성 특징 때문에 설사 증상을 보였음.
- 이 단백질은 기능성 식품의 재료로 유용하게 쓰일 수 있음.

Table 2. Anti-constipation effect of dietary sericin in atropine-treated rats

	Saline		Atropine	
	Control	Sericin	Control	Sericin
Fecal wet wt (g)				
0~6 h	0.25±0.05 ^{1)a2)}	0.25±0.03 ^a	0.12±0.03 ^b	0.27±0.03 ^a
0~12 h	0.42±0.05 ^{ab}	0.46±0.05 ^a	0.28±0.04 ^b	0.48±0.05 ^a
Fecal dry wt (g)				
0~6 h	0.17±0.03 ^a	0.14±0.01 ^a	0.07±0.02 ^b	0.17±0.02 ^a
0~12 h	0.27±0.03 ^a	0.26±0.02 ^a	0.18±0.03 ^b	0.29±0.03 ^a
Fecal water content (%)				
0~6 h	31.7±3.0 ^b	38.9±3.4 ^{ab}	32.9±1.6 ^{ab}	39.6±1.5 ^a
0~12 h	33.5±2.2 ^b	40.0±1.5 ^a	35.4±1.5 ^{ab}	40.4±1.5 ^a

¹⁾Values are means±SE (n=11). ²⁾Within a row, values followed by different letters are significantly different (p<0.05).

Table 3. Anti-colon tumor effect of dietary sericin in 1,2-dimethylhydrazine-treated mice

	Control	Sericin
Mice with colon tumors (n/40)	15/40	5/40*
Tumors/mouse (n)	0.55±0.13 ¹⁾	0.13±0.05*
Proliferative cells (%)	7.92±0.29	5.90±0.32*
<i>c-myc</i> expression cells (%)	7.63±0.24	5.46±0.24*
8-OHdG expression cells (%)	6.58±0.51	4.24±0.24*
4-HNE expression cells (%)	8.16±0.16	5.32±0.12*
iNOS expression cells (%)	9.92±0.62	8.37±0.15*

¹⁾Values are means±SE (n=20). *Significantly different (p<0.05).

Table 4. Mineral absorption-enhancing effect of dietary sericin in rats

Apparent absorption of minerals (%)	Control	Sericin
Zn	37.0±4.3 ¹⁾	52.1±3.6*
Fe	34.4±3.9	48.6±3.0*
Mg	61.6±3.2	74.4±3.3*
Ca	61.0±3.5	71.1±2.2*

¹⁾Values are means±SE (n=6). *Significantly different (p<0.05).

✓ Coimmobilization of Naringinases on Silk Fibroin Nanoparticles and Its Application in Food Packaging⁴

- 생체적합성이 뛰어난 실크 피브로인의 용액을 acetone에 넣어 crystalline silk fibroin nanoparticles(SFNs)로 만들어 glutaraldehyde와 가교를 이뤄 naringinase를 공유결합으로 결합이 형성됨.
- Naringinase는 α -L-rhamnosidase와 flavonoid- β -glucosidase로 구성된 bienzyme으로 55°C에서 8~10 시간 동안 최적 상태가 됨. 뿐만 아니라, 원심분리로 기질과 효소를 분리할 수 있어, 8번 원심분리를 한 후, 그 활성도를 측정함. 그 결과는 원래의 활성도의 70%정도를 보임.
- 이 SFN Naringinase는 적은 비용으로 citrus juice에서 덜 쓰게 만드는 음식 공정에 효과적으로 사용할 수 있음

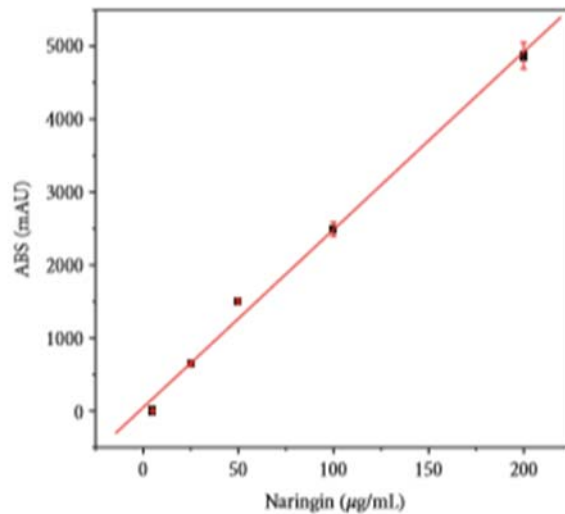


FIGURE 1: Standard curve of naringin by HPLC.

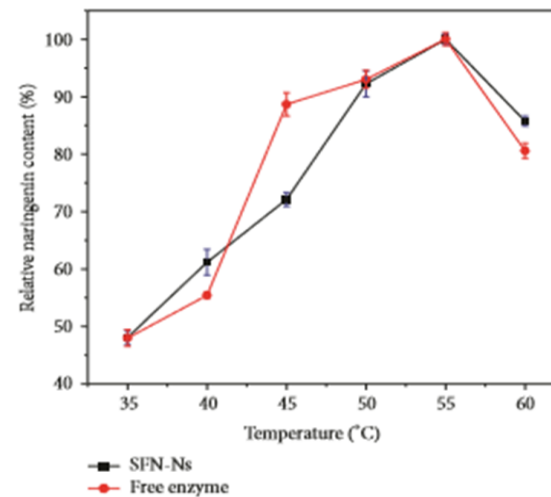


FIGURE 5: Effect of temperature on the hydrolysis degree of naringin by free and SFN naringinases.

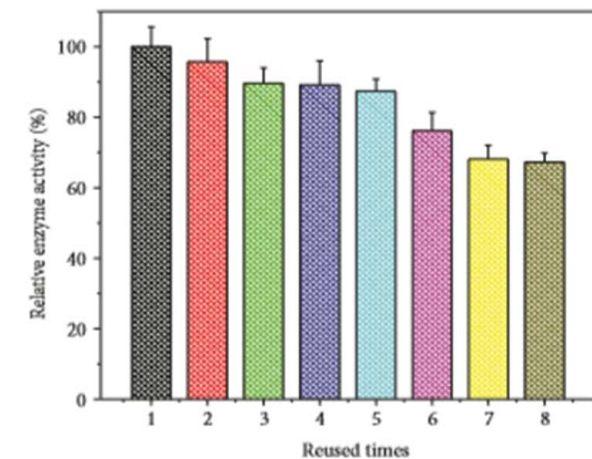


FIGURE 6: Operational stability of the SFN naringinases: the first value in the figure was set as 100%.

✓ Silk Fibroin as Edible Coating for Perishable Food Preservation⁵

- 물에서 Micells나 nanoparticle는 구조적으로 biopolymer로써 생물적 환경과 무생물적 환경을 연결하는 특징을 가지게 됨.
- 실크 피브로인의 전형적인 속성 (다형성(polymorphism), 순응도(conformability) 및 소수성)을 활용하여 딥 코팅시 음식 표면에 자체 조립되는 수용성 단백질 용액을 만듦.
- 코팅된 silk fibroin의 얇은 막이 가스를 확산을 조절할 수 있게 되는데 이러한 원리는 silk fibroin의 많은 beta-sheet 구조가 산소 확산을 감소시킴으로써 음식의 신선도를 유지하게 된다는 것을 보고함.
- 바나나와 딸기에 이러한 코팅을 사용했을 때, cell의 호흡속도와 물의 증발을 감소시킴으로써 상온에서 수확된 과일의 신선도를 오래 유지할 수 있음을 보고함.

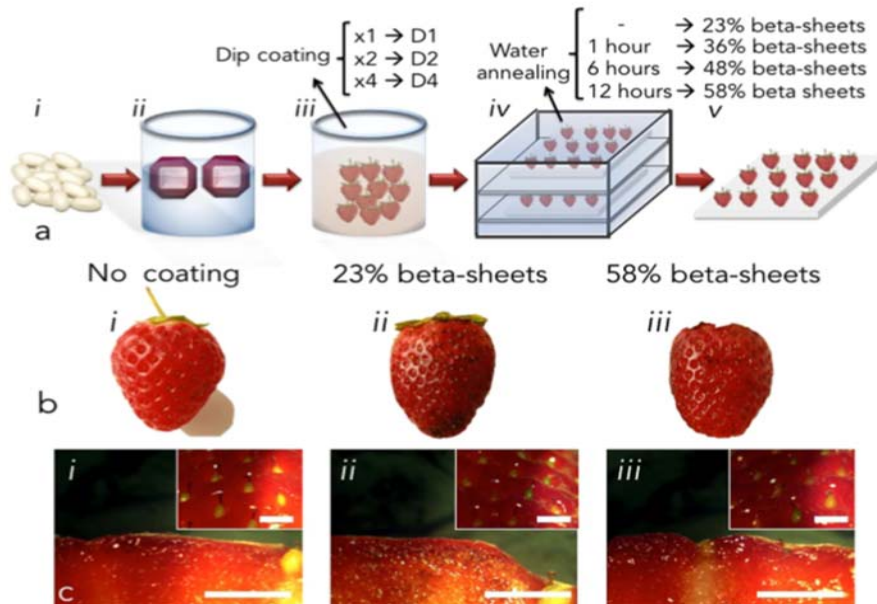


Figure 1. Coating of perishable fruits with edible silk fibroin. (a) The impact of silk fibroin coating was investigated on freshly picked strawberries. (i) Silk fibroin was extracted from *Bombyx Mori* cocoon fibers by dissolution in 9.3 M LiBr solution and (ii) dialysis in deionized water. The concentration of the protein in water was then adjusted to 1 wt%. (iii) Coating of strawberries was then achieved by dip coating process in silk fibroin suspension (1 wt%). The dip coating process was repeated up to 4 times. (iv) Beta-sheet content in silk fibroin edible coatings was modulated using water annealing post-processing. The longer the exposure to water vapour (up to 12 hours), the higher the beta-sheet content of the protein, as reported in Table 1. (v) Silk fibroin-coated strawberries were then left at room conditions ($T = 22^\circ\text{C}$, $\text{RH} = 38\%$) to investigate the impact of the coating on the quality of the fruit. Crystal violet dye was used to stain the silk fibroin coating. (b) Representative images of stained strawberries (i) freshly picked, (ii) coated silk fibroin edible coating (4 dip coating processes, 23% beta-sheet, i.e. no water annealing applied), (iii) coated with silk fibroin (4 dip coating processes), 58% beta-sheet, i.e. exposed to water annealing post-processing. The crystal violet dye is barely visible on the surface of the coated strawberries (black dots) due to the few-micron thickness of the coating. (c) Stereoscopic images of the surface and of the cross-section (insets) of crystal violet-stained fresh strawberries (i) as picked, (ii) coated with silk fibroin with 23% beta-sheet content and (iii) coated with silk fibroin (4 dip coating processes) with 58% beta-sheet content. Scale bars: 2 mm.

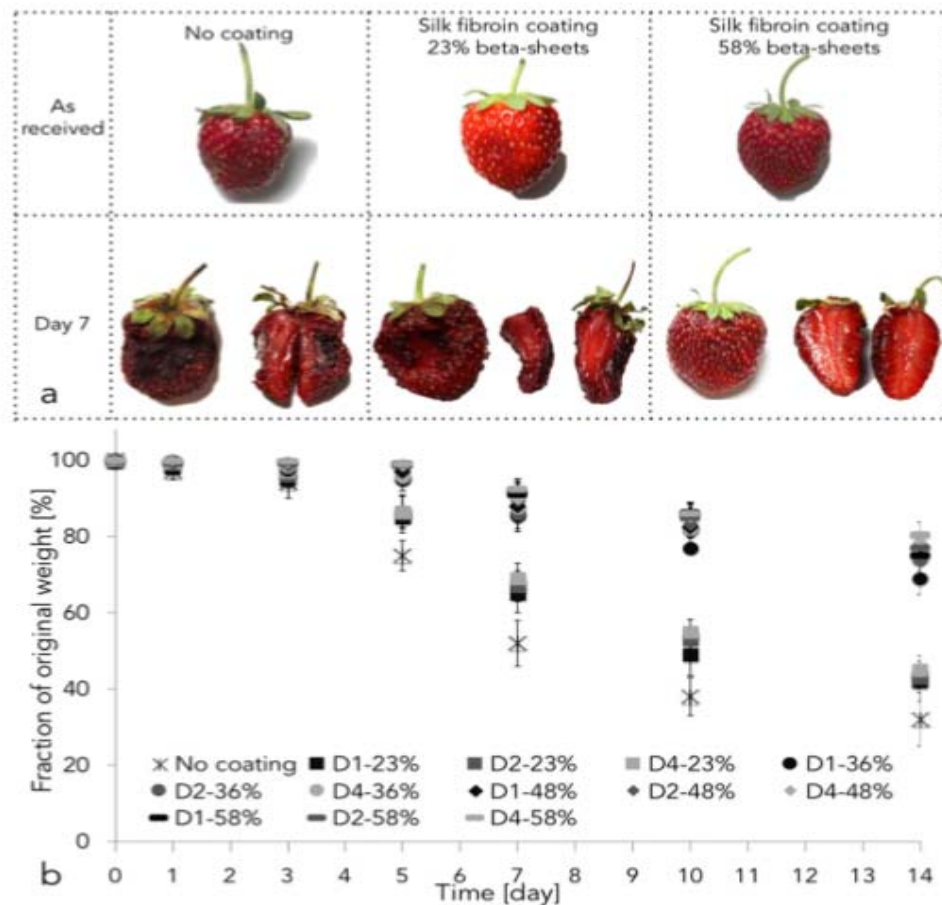


Figure 2. Ripening and weight loss of strawberries coated with edible silk fibroin coating. (a) Time-lapse of strawberries ripening. As picked strawberries were stored at 22 °C and 38% RH (no coating) or dip coated in silk fibroin suspension (23% beta-sheets coating). Water annealing was used as post-process to modulate the relative content of beta-sheets in silk fibroin. At day 7, silk fibroin coating showed to improve the quality of the stored strawberries. (b) Weight loss of strawberries stored for up to 14 days at 22 °C and 38% RH. Strawberries were stored as picked (i.e. *no coating*) or after coating with silk fibroin suspension (*Dx-xx%*). *Dx* stands for 'x' dip coating steps. *xx%* stands for relative amount of beta-sheet content. (e.g. D1-23% means that strawberries were dip coated with silk fibroin materials with 23% beta-sheets content). One-way ANOVA test with Tukey mean analysis was used to evaluate the weight loss data. Silk beta-sheet content (which is proportional to water-annealing time) but not number of dip coating steps affected the dehydration of the strawberries considered. *No coating* controls lost circa 70 wt% of their original weights in the 14 days considered (highlighted within the red rectangles). Strawberries coated silk fibroin retained more water than the *no coating* controls at day 3 ($p < 0.05$). An increase in silk fibroin beta-sheets content via water annealing process further decreased fruit dehydration compared to non-water annealed counterparts ($p < 0.05$) and to the *no coating* controls ($p < 0.05$) but no statistically significant difference was found between samples with a beta-sheet content $\geq 36\%$ ($p > 0.05$).

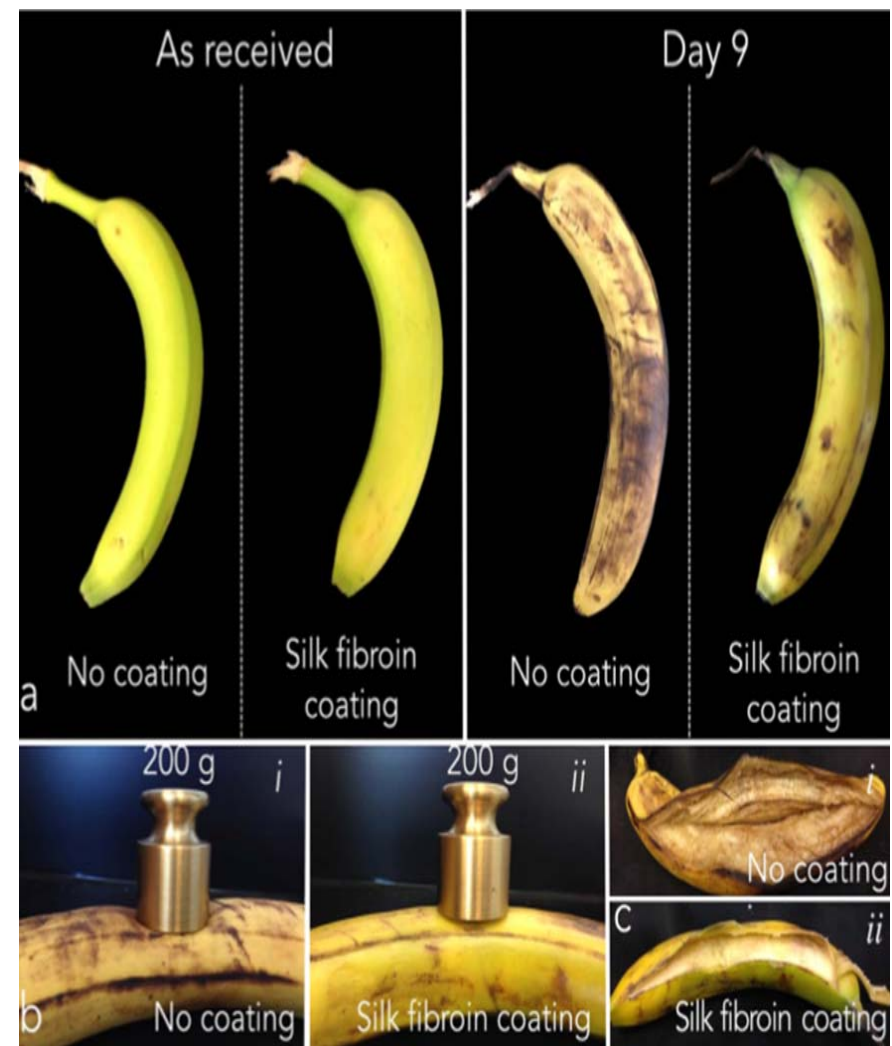


Figure 4. Evaluation of bananas ripening with and without crystalline silk coating. Fruits were stored at 22 °C and 38% RH as received (*no coating*) and after coating with silk fibroin materials. Bananas were hanged on their respective stem throughout the whole experiment. (a) Time lapse photography of banana ripening indicating that silk fibroin coating decreased the ripening rate. (b) Investigation of silk fibroin-coated banana rigidity (i) when compared to uncoated controls. Turgidity was studied qualitatively by applying a dead load (200 g) on the surface of the fruit. (ii). The test was accomplished ad day 9 after coating. Silk fibroin-coated banana showed an increased firmness, when compared to uncoated control. (c) Images of the internal flesh of non-coated (i) and silk fibroin-coated (ii) bananas at day 9 post coating-treatment. Flesh of non-coated banana presented a brown colour, while silk fibroin-coated fruits preserved a tallow flesh, indication of a decreased ripening rate for the silk fibroin-coated samples.

✓ Development of Bread Supplemented with the Silk Protein Sericin⁶

- Silk protein의 하나인 sericin을 빵에 넣었을 때, sericin의 효과적인 섭취를 촉진하고 빵 구조 및 기호성에 미치는 영향에 대해, 구운 빵의 성능을 평가하기 위한 목적으로 sericin 첨가 빵을 개발하여 보고함.
- Sericin을 빵에 넣었을 때, 빵 한 덩이의 크기와 빵 껍질의 어두운 색을 감소되는 것을 확인하였고 다른 물리적 특징이 변하지 않음.
- Sericin을 2g을 넣은 빵과 6g을 넣은 빵을 더 선호하는 경향을 보임. Sericin을 넣음으로써 밀가루의 양은 줄어들었으나, 맛에 큰 변화가 없었음. 빵 한 덩이의 크기와 빵 껍질의 색깔, 감각적인 평가를 통한 선호도 조사의 결과, 가장 적합한 sericin의 양은 2g임을 확인 함.
- Sericin 2g을 넣은 빵을 6개로 잘랐을 때, 한 개의 slice의 빵에는 330 mg의 sericin을 들어가게 됨. 330mg의 sericin에는 약 20% 저항 단백질이 생기게 되므로, 한 개의 slice의 빵에는 약 67mg의 저항 단백질이 생기게 됨.
- Sericin을 넣은 빵이 sericin을 효율적으로 섭취하는 적합한 가공 음식임을 보고함.

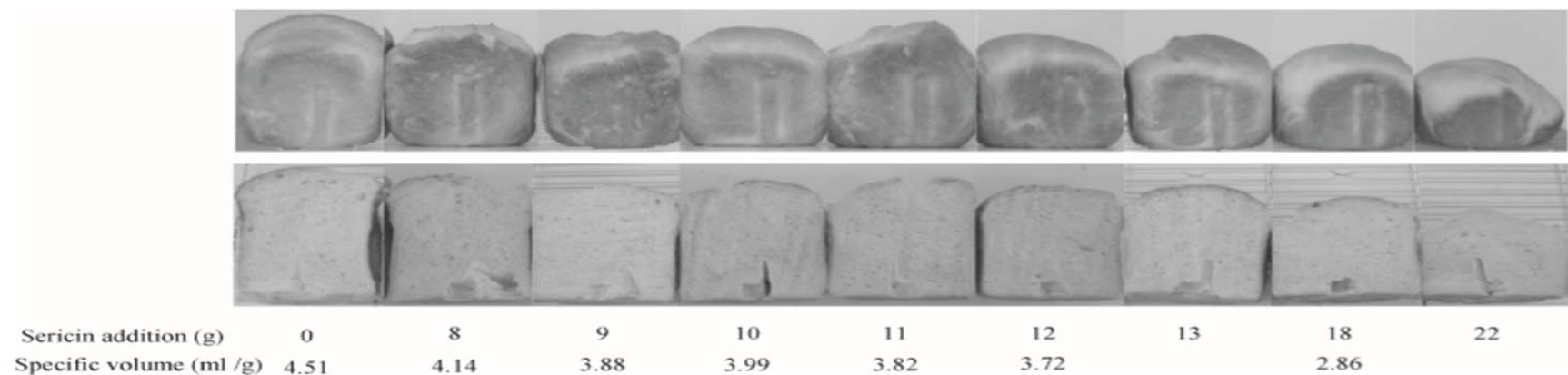


Fig. 1. Images of breads and cross-sections of loaves prepared with various sericin additions

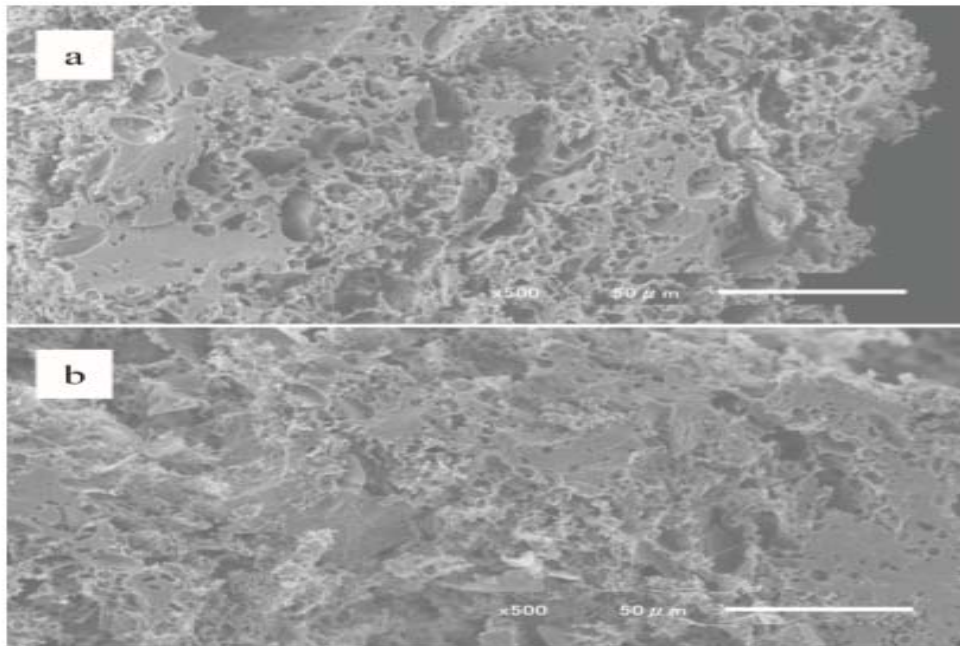


Fig. 2. SEM image of model dough with (b) or without (a) sericin

Table 4. Effects of added sericin on bread palatability

	Sericin addition (g)	crust color	texture	hardness	flavor	chewiness	ease of swallowing	overall evaluation
	0 (control)	0	0	0	0	0	0	
Discrimination	2	0.03 ± 1.20	-0.46 ± 1.24	-0.17 ± 1.33	-0.37 ± 1.00	0.14 ± 1.12	-0.20 ± 0.90	
	4	0.40 ± 1.03	0.06 ± 1.11	0.34 ± 1.19	-0.20 ± 1.05	0.17 ± 1.12	0.20 ± 1.08	
	6	0.26 ± 1.29	0.23 ± 1.21	0.60 ± 1.42	-0.06 ± 1.35	0.00 ± 1.24	-0.63 ± 0.97*	
	8	0.77 ± 1.11*	0.34 ± 1.26	0.71 ± 1.27*	-0.03 ± 1.42	-0.26 ± 1.33	-0.40 ± 1.22	
	0 (control)	0	0	0	0	0	0	0
Preference	2	0.26 ± 0.98	-0.03 ± 1.27	-0.11 ± 1.37	0.29 ± 0.99	0.29 ± 1.20	0.26 ± 0.95	0.46 ± 1.01*
	4	0.46 ± 1.07	0.34 ± 1.08	-0.20 ± 1.16	-0.06 ± 1.11	0.11 ± 1.23	0.06 ± 1.03	0.20 ± 0.93
	6	0.40 ± 1.06	0.09 ± 0.98	0.40 ± 1.35	-0.11 ± 1.08	0.20 ± 1.02	-0.09 ± 0.92	0.06 ± 1.21
	8	0.06 ± 1.21	-0.09 ± 1.31	0.43 ± 1.44	-0.34 ± 1.21	0.29 ± 1.30	-0.34 ± 1.28	-0.23 ± 1.17

n = 35, mean ± SEM; *:p < 0.0125

3. 결론

- ✓ 4차 산업 혁명시대를 맞이 하면서 다양한 소재 개발과 이에 따른 산업의 응용성이 매우 중요해 지고 있음.
- ✓ SF의 응용으로 건강 기능성 제품, 식품 소재 개발, 실크 화장품, 실크 비누등의 인체 친화용 소재 개발, 생체 의료용 소재 개발, 부직포나 섬유 가공제 등의 산업용 소재로서 부가가치가 기대됨.^{1,2}
- ✓ 실크 피브로인의 그 기능과 구조에 따라 체지방 감소, 인슐린 분비촉진, 간 기능 개선 및 알코올 흡수촉진, 퇴행성 신경질환개선 및 예방(치매, 알츠하이머), 기억력 및 인지능력 향상, 집중력 향상, 칼슘 흡수 촉진, 혈중콜레스테롤 저하 촉진 등으로 구분되어 특정 질병의 치료제와 같은 의약학적인 용도로 기술 개발이 보고 되고 있음.⁶
- ✓ 향후 실크 펩타이드의 활성화에 대한 규명이 명확해 진다면 새로운 생리 활성을 갖는 신 물질이 개발될 수 있을 것이라 판단됨.⁶

4. 참고 문헌

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