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Improvement of Atopic Dermatitis by Human Sebaceous Fatty Acids and Related Lipids

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1. Introduction

Our bodies are protected against a variety of external factors such as transient pathogens and can preserve homeostasis. This defense mechanism consists of many components, including antimicrobial lipids such as *cis*-6-hexadecenoic acid (C16:1Δ6) (Wille & Kydonieus, 2003) and sphingosine (Arikawa et al., 2002), as well as antimicrobial peptides such as β-defensins and cathelicidin (Harder et al., 2010; Hsu et al., 2009; Tay et al., 2011). The levels of these antimicrobial components in the skin of atopic dermatitis (AD) patients are lower than those of healthy subjects (Arikawa et al., 2002; Harder et al., 2010; Takigawa et al., 2005). It has been reported that the uncommon lipid C16:1Δ6 is present as a major component of sebaceous lipids in human skin and hair but that it is not generally included in plant or animal oils. We have focused on this very unique unsaturated fatty acid, and have been investigating the relationship between this fatty acid and the resident flora of human skin as well as transient pathogens. In the course of that research, we found a correlation between C16:1Δ6 and AD, and have developed a method for the production of C16:1Δ6 on an industrial-scale (Araki et al., 2007; Koike et al., 2000a, 2000b; Takeuchi et al., 1990). In addition, we have made further progress in antimicrobial research involving C16:1Δ6 (Araki et al., 2005) and have selected fatty acid derivatives (oxa-fatty acids) with higher activity by means of analyses of structure-activity relationships (Sugai et al., in preparation).
In this chapter, we introduce our research into fatty acids, and we also review approaches to normalizing AD from a microbiological point of view.

2. Function of human sebaceous fatty acid "C16:1Δ6"

Human skin lipids, including sphingosine and C16:1Δ6, provide a defense against external factors and function to maintain homeostasis. It is well established that human skin lipids contribute to the defense mechanism termed "self-sterilization" (Wille & Kydonieus, 2003) and the antimicrobial activities of each lipid component have been evaluated (Arikawa et al., 2002; Harder et al., 2010). A significant difference was found in the fatty acid composition of sebum lipids between AD patients and healthy controls (Takigawa et al., 2005). We describe the results of our investigation in the section below.

2.1 *Cis*-6-hexadecenoic acid (C16:1Δ6)

Cis-6-hexadecenoic acid, termed sapienic acid, is unique in the animal kingdom, being a 16-carbon mono-unsaturated fatty acid with a *cis*-double bond located at the sixth carbon from the carboxyl terminal (Fig. 1).

Fig. 1. Structure of *cis*-6-hexadecenoic acid (C16:1Δ6)

This fatty acid is an isomer of palmitoleic acid (*cis*-9-hexadecenoic acid (C16:1Δ9)), which is a common constituent of the glyceride of human adipose tissue. In addition, C16:1Δ6 is a major component of sebaceous wax esters, triacyl glycerols, and of free fatty acids found in skin and hair (Downing & Strauss, 1974; Morello & Downing 1976; Yamamoto et al., 1987). Sapienic acid represents 85% of the lipid in *Thunbergia alata* seed oil, and is obtained either by extraction of this plant or by chemical synthesis (Spencer et al., 1971).

2.2 Relationship between human sebaceous fatty acid C16:1Δ6 and AD

The composition of free fatty acids in the sebum at the recovery level is shown in Figure 2 (Takigawa et al., 2005). The level of C16:1Δ6 in healthy control skin ranged from 0.43 to 5.38 μg/cm² with a mean of 2.0 μg/cm² (4.9 to 16.8% with a mean of 12.9% in percent of total)

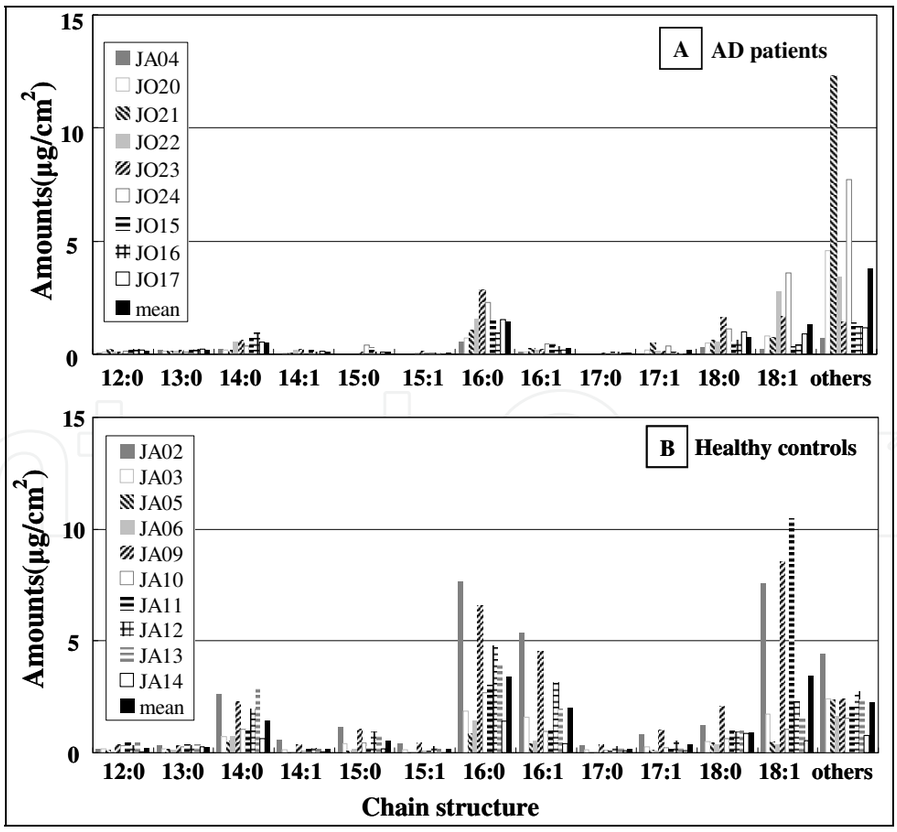

Fig. 2. Amount of free fatty acid in sebum from AD patients (upper panel) and from healthy controls (lower panel). Sebum was analyzed at the recovery level.

and the total level of C16:0 ranged from 0.88 to 7.64 μg/cm² with a mean of 3.43 μg/cm² (14.9 to 28.2% with a mean of 22.1% in percent of total). In contrast, the level of C16:1Δ6 (0.08 to 0.53 μg/cm² with a mean of 0.26 μg/cm²) and the percent of total (1.1 to 8.7% with a mean of 3.0% in percent of total) in the skin of AD patients were lower than in the healthy controls, although the amounts of C16:0 (0.53 to 2.84 μg/cm² with a mean of 1.44 μg/cm²) and the percent of total (6.8 to 30.7% with a mean of 16.5% in percent of total) were similar to those of the healthy controls. A significant decrease in free C16:1Δ6 content in nonlesional skin from AD patients compared with healthy controls was found.

2.3 Antibacterial activity of C16:1Δ6: Selective antibacterial activity

Colonization of *S. aureus* has been reported in AD patients and is regarded as an exacerbation factor (Akiyama et al., 1996). Antimicrobial agents, including C16:1Δ6, inhibited the growth of *S. aureus* at a final concentration of 100 mg/l in the following order; Benzalkonium chloride = Triclosan > C16:1Δ6 > Trichlorocarbanilide, while they inhibited the growth of *S. epidermidis* in the following order; Benzalkonium chloride = Triclosan> C16:1Δ6 = Trichlorocarbanilide (Fig. 3). Whereas C16:1Δ6 could inhibit the growth of *S. aureus* over a concentration range from 100 to 1000 ppm, it did not greatly inhibit *S. epidermidis*.

In addition, the antibacterial mechanism of C16:1Δ6 against *S. aureus* and *S. epidermidis* was investigated using protoplasts treated with lysostaphin. The results of this experiment indicated that there was no difference in the effectiveness of C16:1Δ6 against *S. aureus* and *S. epidermidis*, and suggested that the antibacterial action of C16:1Δ6 against Staphylococcal bacteria was caused by disruption of the cell membrane, not disruption of the cell wall, and that this disruption resulted in leakage of bacterial components (Data not shown).

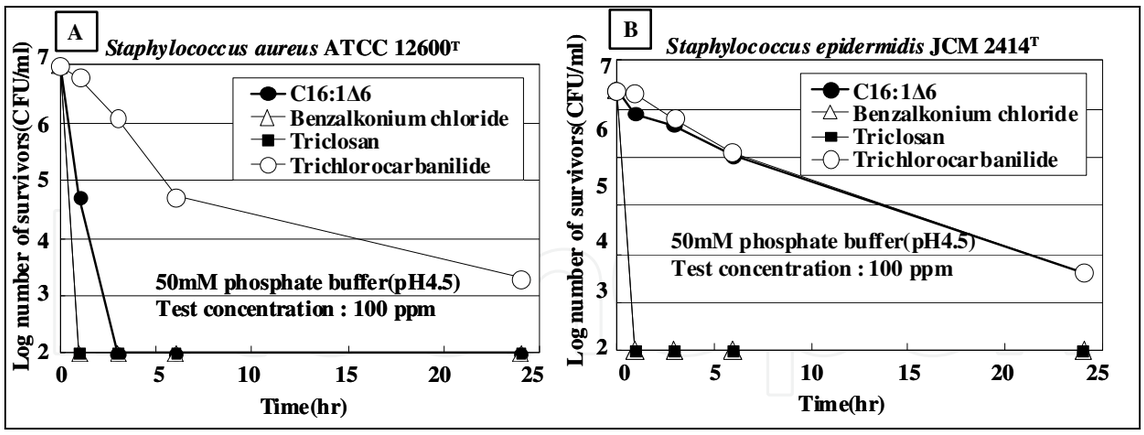

Fig. 3. Comparison of the antimicrobial effects of C16:1Δ6 against *Staphylococcus aureus* ATCC 12600T (left panel) and *Staphylococcus epidermidis* JCM 2414T (right panel).

2.4 Topical application of C16:1Δ6 to AD patients

An inverse correlation between *S. aureus* colonization and the level of C16:1Δ6 in AD patients was found (Fig. 4). In addition, the colonization of *S. aureus* was reduced by topical application of C16:1Δ6 to the skin of six to eight AD patients in a small-scale clinical study. Based on these investigations, it was considered that the decreased level of

C16:1Δ6 in AD patients may be one of the factors that contribute to the colonization of *S. aureus* (Takigawa et al., 2005).

We have confirmed that C16:1Δ6 functions as a natural antibacterial component and has very unique properties including "selective antibacterial activity", in which it shows effective antibacterial activity against transient *S. aureus* but not against residential *S. epidermidis*. By topical application of this fatty acid to AD patients, the colonization of *S. aureus,* which is believed to be an exacerbation factor for this disease, was repressed, thereby indicating the effectiveness of this fatty acid. From the results of these investigations, a new approach for improvement of skin disorders, based on normalization of the microflora on human skin, was proposed using the selective antibacterial activity of C16:1Δ6.

3. Proposed industrial production of C16:1Δ6 using a bioreactor

Microbial production of various materials has a number of advantages over chemical synthesis such as cost effectiveness, ease of production and regiospecific production. Considering the unique function of C16:1Δ6, and its potential therapeutic usefulness, we investigated the possibility of microbiologically producing C16:1Δ6.

There are many reports regarding microbial production of polyunsaturated fatty acids under simple and mild conditions. The production of γ-linoleic acid and arachidonic acid by species of the fungus *Mortierella* is a valuable product for many applications, and the triglyceride esters of poly unsaturated fatty acids that are produced in a fermentation process are used in skin care products (Certik & Shimizu, 1999a; Suzuki, 1987, 1988; Yamada et al., 1987) and baby formula (Certik & Shimizu, 1999b; Shinmen et al., 1989; Yamada, 1988). We isolated an alkane-assimilating *Rhodococcus* sp. strain from a soil sample and found that a mutant was capable of introducing a *cis*-double bond into various aliphatic substrates. We used this mutant for production of C16:1Δ6, investigated the production process, and further constructed a bioreactor system for its industrial production.

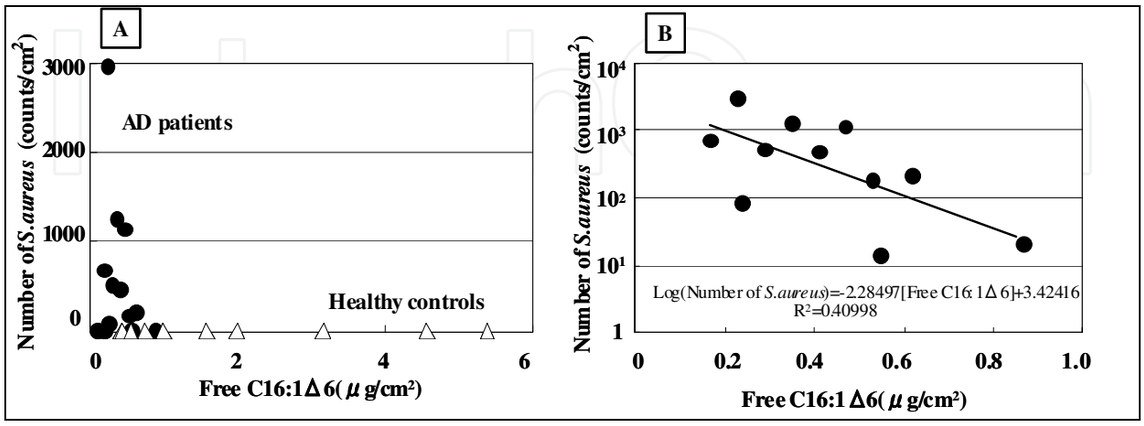

Fig. 4. Relationship between free C16:1Δ6 and *S. aureus* in the skin. A: in the skin of AD patients and Healthy Controls; B: in the skin of AD patients who exhibited more than 10 counts/cm² of *S. aureus*. AD patients; (●), Healthy Controls; (Δ)

3.1 Desaturation reaction of aliphatic substrates by a mutant strain of alkane-assimilating *Rhodococcus* sp.

We found that the double mutant designated *Rhodococcus* sp. KSM-MT66 had the ability to desaturate various aliphatic compounds such as alkanes, chloro alkanes and fatty acid esters, and that this mutant strain produced unsaturated compounds extracellularly. When hexadecanoic acid esters such as methyl, propyl, isopropyl, and isobutylester were supplied as substrates to the resting cells, their corresponding *cis*-desaturated compounds were produced at a concentration of 0.5, 20, 53 and 7 g/l in 3 days. The enzyme(s) responsible for the desaturation reaction appears to recognize mainly the sixth carbon from the carbonyl carbons (Fig. 5). The bioproduction of C16:1Δ6 esters has not been previously reported (Koike et al., 1999). When alkanes were supplied to this mutant strain, the main products had a double bond at the ninth carbon from the terminal methyl group. These experiments indicated that the unsaturated position differed according to differences in the supplied substrates (Koike et al., 2000a).

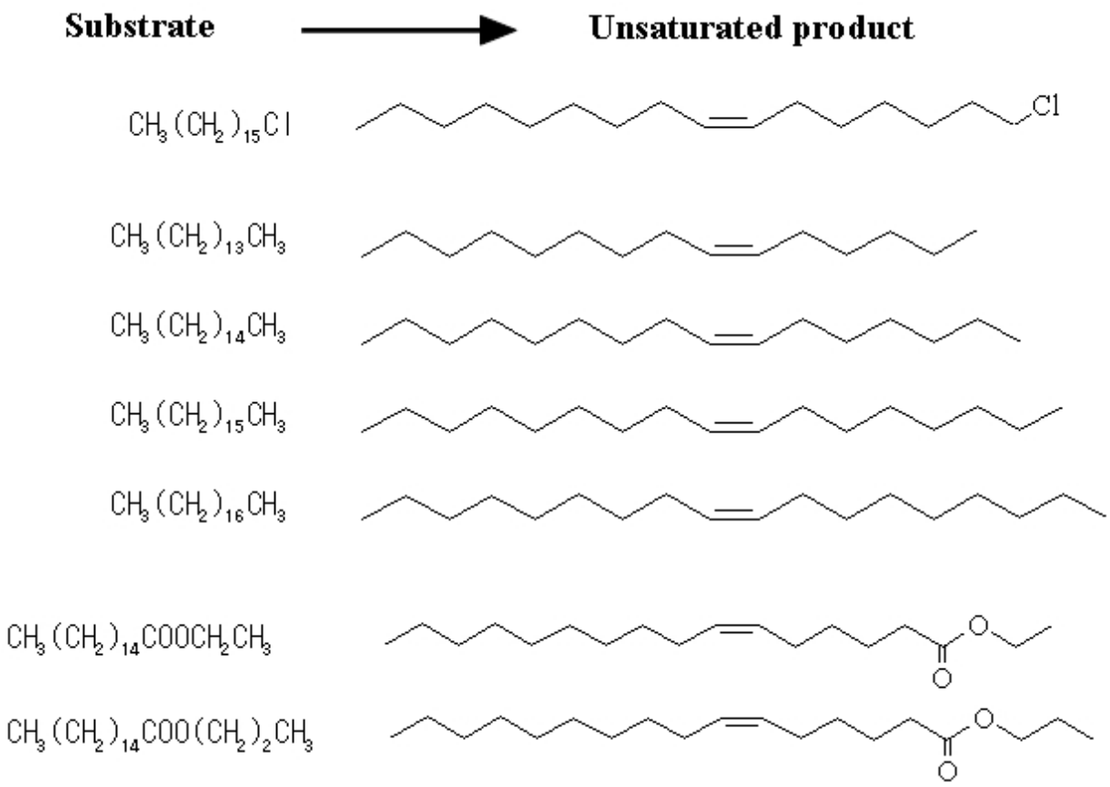

Fig. 5. The patterns of regiospecific desaturation of aliphatic substrates by *Rhodococcus* sp. strain KSM-MT66 cells.

3.2 Production of a C16:1Δ6 ester by resting cells of *Rhodococcus* sp. KSM-MT66

We determined the condition with resting cells of the *Rhodococcus* sp. KSM-MT66 strain to produce C16:1Δ6. The reaction mixture contained 20% (w/v) hexadecanoic acid isopropyl ester (IP-C16:0), 0.25 M phosphate buffer (pH 7.0), 1.0% (w/v) monosodium glutamate, 2

mM thiamine, 2 mM $MgSO_4$, and 5% (wet w/v) resting cells. Bioconversion was performed using a 1.0 l working volume in a 2.6 l bioreactor at 26 °C with aeration and agitation. Glutamate, thiamine, and $MgSO_4$ prevented cell damage of this strain during repeat-batch bioconversion, and optimum concentrations of these factors were maintained in the reaction mixture. Under optimum conditions, about 50 g/l of C16:1Δ6 isopropyl ester (IP-C16:1Δ6) was produced extracellularly in 3 days.

Since this reaction mixture consisted of water, oil and cells, we considered that this may make it difficult to separate the products. We therefore designed a new process to recover the products. Thus, the oil in water (O/W-type) emulsified reaction mixture was kept without agitation for 20 hours, the water layer was drained out, an appropriate volume of substrate, IP-C16:0, was added to the reaction mixture to invert the emulsion phase to a W/O-type, and the product, IP-C16:1Δ6, in the continuous oil phase was recovered through a hydrophobic hollow-fiber module (Fig. 6). The hydrophobic cells were concentrated in the oil phase. It was then possible to start the next batch by adding fresh medium to these cells (Koike et al., 2000b; Takeuchi et al., 1990). This system allows repeat-batch reactions without having to use an organic solvent to recover the products.

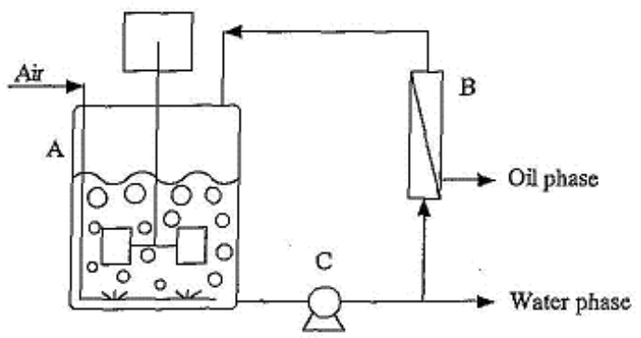

Fig. 6. Schematic diagram of the membrane bioreactor system for Rhodococcal bioconversion. (A) Reactor vessel; (B) Hydrophobic hollow-fiber module; (C) Circulation pump.

3.3 Repeat-batch production of the C16:1Δ6 ester

The reason for the decline in late production during repeat batch production may be due to various factors such as accumulation of some inhibitory products, cell damage and/or exhaustion of some nutrients. Since monosodium glutamate was found to reduce cell damage, we investigated the effect of monitoring and adjusting the concentration of monosodium glutamate on productivity during repeat batch production (see sec. 3-2). The results of this experiment showed that a productivity of 0.8 g/l was attained over the course of 13 cycles (300 hours) by maintaining the concentration of monosodium glutamate between 0.5 and 1.5% (Fig. 7). It was possible to recover the produced IP-C16:1Δ6 using the hollow-fiber system, and IP-C16:1Δ6 was then purified by urea adduct treatment, evaporated and applied to a silica gel column. The total yield of IP-C16:1Δ6 using this procedure was 79% and the purity was over 97%. Long term operation of C16:1Δ6 ester production was achieved using a 2.6 l fermentor (Koike et al., 2000b).

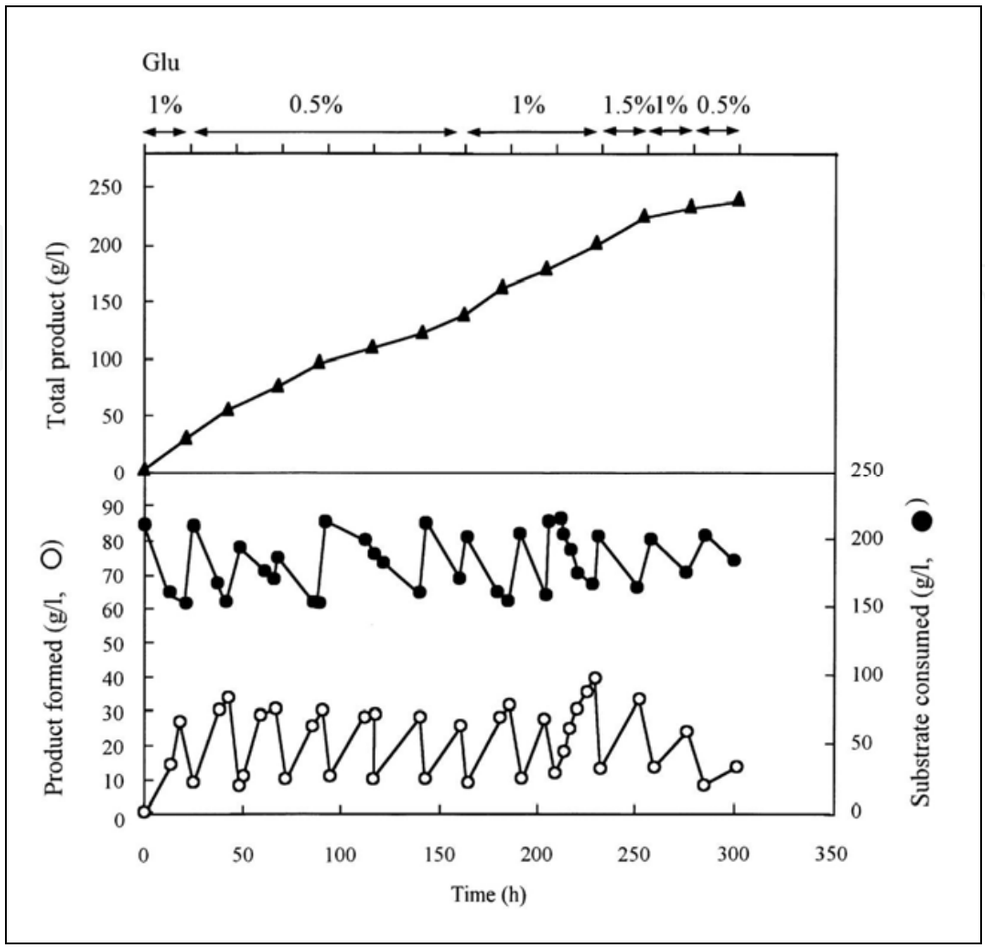

Fig. 7. Repeat-batch reaction using a membrane bioreactor system with a phase inversion for the production of IP-C16:1Δ6 by the resting cells of *Rhodococcus* sp. KSM-MT66.

3.4 Fermentative production of IP-C16:1Δ6 using growing cells of the mutant *Rhodococcus* sp. KSM-MT66

The resting cell production of IP-C16:1Δ6 described above includes four complicated steps; (i) cell growth, (ii) cell harvesting, (iii) incubation of cells with substrate and (iv) phase-inversed separation of the product using a hydrophobic fiber membrane system.

In order to develop a more convenient production process we therefore investigated production of IP-C16:1Δ6 using a fermentation process. We first established a basal medium for *Rhodococcus* sp. KSM-MT66. When production was performed under resting cell conditions, 18 g/l of IP-C16:1Δ6 was produced over 3 days of cultivation. Optimization of the concentrations of metal ions greatly improved production of IP-C16:1Δ6, resulting in production of 52 g/l over 4 days of cultivation (see sec. 3.5).

3.5 Fermentative production of IP-C16:1Δ6 using the mutant *Rhodococcus* sp. KSM-T64

Since there was a possibility that the IP-C16:1Δ6 product might be degraded by esterases present in the *Rhodococcus* sp. KSM-MT66, we attempted to create *Rhodococcus* mutants with

reduced esterase activity by UV irradiation. Of the colonies which showed lower growth than KSM-MT66 on the minimum agar containing IP-C16:0, one mutant, designated KSM-T64, displayed 40% of the esterase activity of KSM-MT66.
Using this mutant strain T64, and optimizing culture conditions, more than 60 g/l of IP-C16:1Δ6 could be produced in a flask (Table 1). Optimization of culture conditions in a 30 l jar fermentor, resulted in production of 50 g/l over 4 days of cultivation (Fig. 8). Furthermore, C16:1Δ6 can be easily obtained by simple hydrolysis of IP-C16:1Δ6 (Araki et al., 2007).

Reaction condition			**KSM- MT66**	**KSM-T64**
Reaction process			Resting	Growing
Reaction ingredient	Monosodium glutamate		1.0%	2.0%
	Metal	$MgSO_4 \cdot 7H_2O$	2 mM	2 mM
		$MnSO_4 \cdot 6H_2O$	-[a]	2 μM
		$FeSO_4 \cdot 7H_2O$	-	60 μM
		$CuSO_4 \cdot 5H_2O$	-	5 μM
		$ZnSO_4 \cdot 7H_2O$	-	-
	Thiamine		2 mM	-
	Yeast extract (P-21)		-	0.8%
	Phosphate buffer		250 mM	350 mM
			pH 7	pH 7.3
	IP-C16:0		20%	22%
IP-C16:1 productivity			50 g l^{-1}	61 g l^{-1}

a Not added.

Table 1. Production of IP-C16:1Δ6 by *Rhodococcus* sp. KSM-MT66 and KSM-T64.

4. The production of C16:1Δ6 by genetically modified bacteria: Cloning and expression of two novel desaturases from *Rhodococcus* sp.

4.1 Review of desaturase studies

An enzyme that removes hydrogen atoms from a fatty acid derivative, thereby creating a double bond, is called a desaturase, and desaturases are key enzymes for the maintenance of cell life cycles. Desaturases are divided into three types based on substrate specificity; acyl-CoA, acyl-ACP, and acyl-lipid types (Fox et al., 2004). Desaturases are further sub-divided into membrane-bound and soluble types. Membrane-bound enzymes are very unstable and are known to be very difficult to purify. However, recently a very interesting report has been published regarding the purification of human stearoyl-CoA desaturase (Gorden & Fox, 2008). In contrast, there have been some reports regarding the purification and clarification of the crystal structure of the soluble types of enzymes **(**Lindqvist et al., 1996).

Delta 6-desaturases have been reported to be obtained from animals, plants, fungi and cyanobacteria (Aki et al., 1999; Cahoon et al., 1994; Inagaki et al., 2003; Okayasu et al., 1981; Reddy & Thomas, 1996; Zhan et al., 2004). Human and rat fatty acid desaturase 2 (FADS2) -encoding delta 6-desaturases recognize a saturated fatty acid as a substrate under some conditions (Ge et al., 2003; Guillou et al., 2003), and a human delta 6-desaturase gene has been reported to be expressed only in the sebaceous gland. Regarding microbial delta 6-desaturases, industrial producers of unsaturated fatty acids have focused mainly on fungi of the *Mortierella* spp., especially *Mortierella alpina* and *Mortierella cincinelloides*. Two types of delta 6-desaturases have been reported to be expressed by *Mortierella alpina.* The corresponding genes were isolated and the sequences and functions of these genes have been analyzed (Huang et al., 1999; Sakuradani et al., 1999; Sakuradani & Shimizu, 2003). However, even though many delta 6-desaturase genes have been obtained, no delta 6-desaturase genes have been purified from *Rhodococcus* sp.
We aimed to obtain delta 6-desaturase genes from the *Rhodococcus* sp. KSM-T64 strain, which shows 40% of the esterase activity of the parent strain, KSM-MT66.

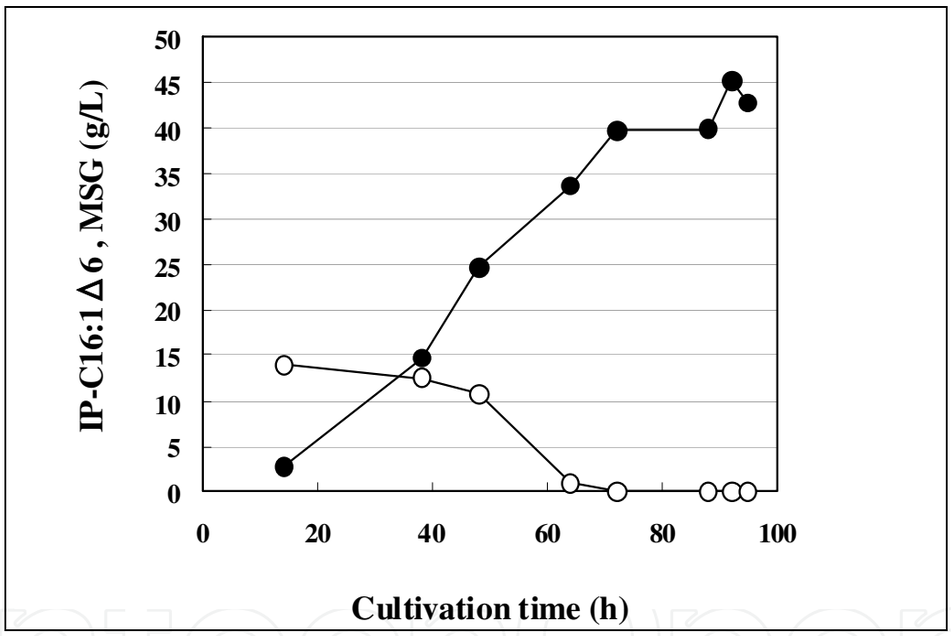

Fig. 8. Production of IP-C16:1Δ6 in a 30 l-jar fermentor under optimized conditions.

IP-C16:1Δ6 (●), monosodium glutamate (MSG) (○). Cultivation was performed at 26 °C, with agitation at 350 rpm, aeration at 0.3 vvm and pressure at 0.2 kg/cm^2. The *Rhodococcus* sp. KSM-T64 was used.

4.2 Cloning of delta 6-desaturase genes from the *Rhodococcus* sp. KSM-T64 strain

The production of IP-C16:1Δ6 by *Rhodococcus* sp. was improved by the addition of metal ions as mentioned above. We therefore targeted the membrane-bound delta 6-desaturase and attempted to clone a gene whose encoded protein could introduce a *cis*-double bond**.** We cloned the delta 6-desaturase gene on the basis of previously reported conserved sequences. Histidine motifs (Shanklin & Fox, 1994; Shanklin, 2009) that bind to ferric ions and whose sequences are known to be conserved in membrane-bound desaturases in a

number of species including worms, borages, *Mucor* sp. and *Mortierella* sp., were targeted for primer design for PCR amplification (Fig. 9). These primers were used for degenerated PCR and nested PCR. In addition, inverse PCR was performed using the obtained PCR products. Approximately 5.5 kbp of sequence was obtained using these methods. Within this sequence, there were sequences that corresponded to two tandem desaturase-like proteins; the first ORF (Rdes1) was comprised of 420 amino acids and the second ORF (Rdes2) was comprised of 413 amino acids from inffered start codons individually. The estimated molecular mass of the Rdes1 protein was 47985 Da and that of Rdes2 was 46951 Da. Three histidine motifs that are established consensus sequences in desaturase proteins were found in these ORF sequences (Fig. 10).

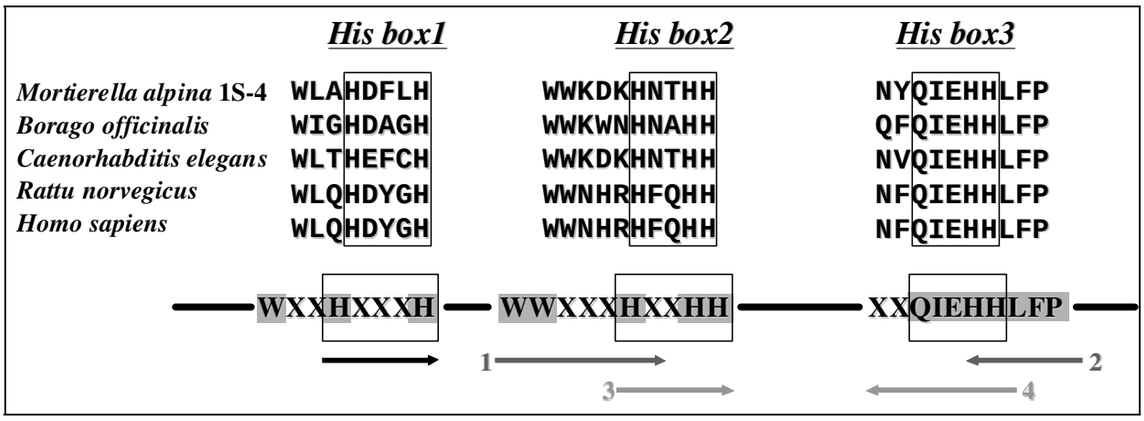

Fig. 9. Construction of PCR primers based on the histidine motifs that are conserved among delta 6-desaturases obtained from various organisms.
The sequence of primers 1, 2, 3, and 4 was; 5′-YTGGTGGAAGGRYAABCAYAA-3′, 5′-RGGGAAVARRTGGTG-3′, 5′-CAYAAYNMDCAYCA-3′, and 5′-RGGGAAVARRTGGTG-3′ respectively. Y, R, B, N, M, D, V, and W were mixed primers of C/T, AA/G, C/T/G, A/C/G/T, A/C, A/G, A/C/G and A/T respectively.

Among the membrane-bound delta 6-desaturases which were clarified the function except for putative and probable delta 6-desaturases, Rdes1 and Rdes2 had about 30% to *Homo sapiens* as an amino acid sequence. An unknown ORF whose function has not yet been clarified is present upstream of these two delta 6-desaturase genes. A domain search revealed that this ORF had an oxidoreductase NAD-binding domain, an oxidoreductase FAD-binding domain, a 2Fe-2S iron·sulfur cluster domain and a ferric reductase domain (data not shown).

4.3 Expression of delta 6-desaturase genes in *Escherichia coli*

Plasmids incorporating the delta 6-desaturase genes Rdes1 and Rdes2 were individually introduced into competent *E. coli* BL21 (DE3) cells, and the corresponding proteins were produced by IPTG induction. Production of very high amounts of these proteins was confirmed using SDS-PAGE. The molecular mass of each of these expressed proteins was 45 kDa. These proteins were identified by comparison with a control cell lysate which was prepared from cells transformed with vector alone.

Fatty acids extracted from *E.coli* transformants that expressed Rdes1 and Rdes2 were analyzed using GC and GC/MS. The level of C16:1Δ6 was increased 2.5-fold in the Rdes1-expressing transformant and 4.8-fold in the Rdes2-expressing transformant, over that of control.

Although the level of C16:1Δ6 fatty acids in the transformants was higher than that of control, it was unclear if these delta 6-desaturase genes of the *Rhodococcus* sp. produced extracellular IP-C16:1Δ6 or not (Araki et al., 2005).

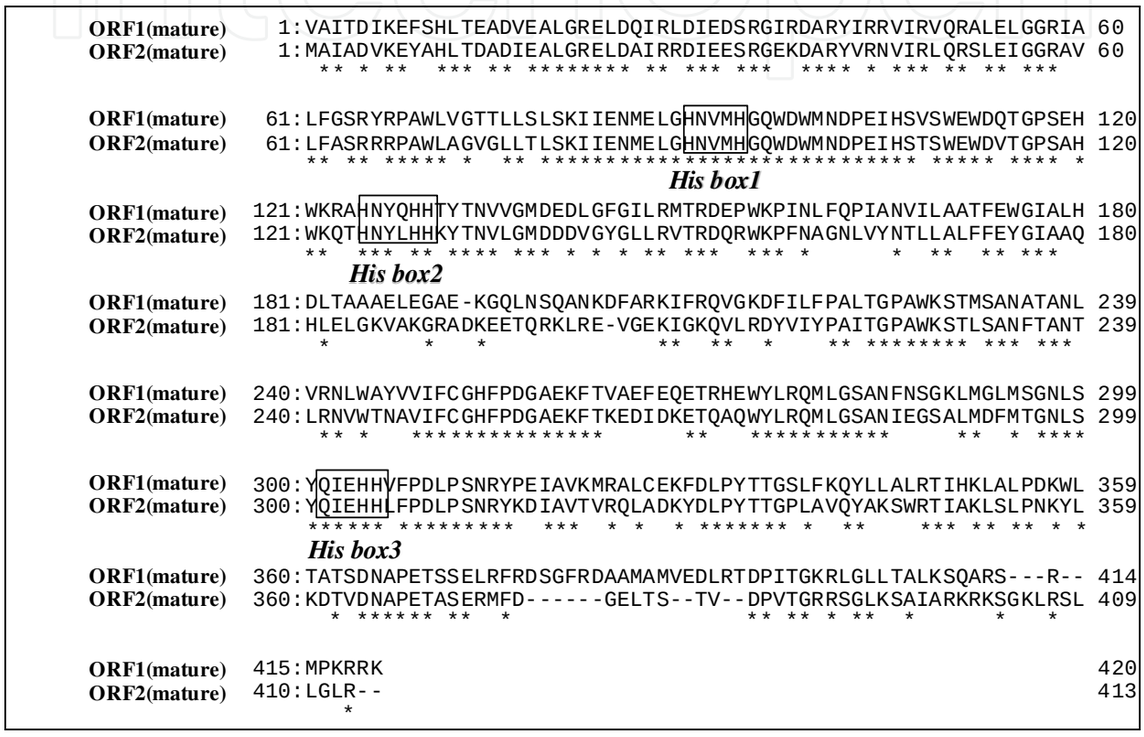

Fig. 10. Sequence homology of the proteins encoded by Rdes1 (ORF1) and Rdes2 (ORF2).

The homology search was performed using Genetyx Win ver.6.1. The gap penalty score was set at –10 as an insert and –3 as an extend. The three histidine rich regions (Histidine motifs) that are conserved in membrane binding desaturases are boxed.

5. Further investigation of human sebaceous fatty acids combined with chemical synthesis: Structure-activity relationship of oxa-fatty acids

C16:1Δ6 has a double bond and, consequently, its stability is a concern, which raises doubts regarding adequate stability of C16:1Δ6 in products. The stability of C16:1Δ6 can be improved by replacing the unsaturated fatty acids with the corresponding oxa-fatty acids (Alkoxy fatty acids). Such a change would also be expected to lead to an improvement in antimicrobial properties of the fatty acid. We describe the results of our investigations regarding such a replacement in the section below.

5.1 Review of the antimicrobial activities of oxa-fatty acids

Although oxa-fatty acids are widely used as intermediate chemicals in fine chemical fields such as medicine, agriculture and fragrance, there have only been a few reports of the antimicrobial activity of oxa-fatty acids. N-alkoxyacetic acid and its methyl ester showed good antifungal activity against *Aspergillus niger*, *Myrothecium verrucaria*, and *Trichoderma viride* (Gershon et al., 1979). In addition, 4-oxatetradecanoic acid, which is an inducer of myristic acid β-oxidation, displayed antifungal activity against *Cryptococcus neoformans* and also antiviral activity against human immunodeficiency virus I (Langner et al., 1992). These studies indicate that oxa-fatty acids of C16:1Δ6, in which the double bond has been replaced with an ether bond (7-oxaheptadecanoic acid or 6-decyloxy hexanoic acid), will maintain their antibacterial activity and will also have a novel function, that is antifungal activity.

5.2 Synthesis and evaluation of a C16 oxa-fatty acid

An oxygen-containing analog of C16:1Δ6 (oxa-fatty acid:7-oxaheptadecanoic acid) was synthesized by condensation of *n*-decanol with 6-bromo-hexanoic acid under alkaline conditions. The antimicrobial activities of purified 7-oxaheptadecanoic acid were then compared with those of C16:1Δ6. The oxa-fatty acid analog of C16:1Δ6, 7-oxaheptadecanoic acid, retained the selective antimicrobial activity of C16:1Δ6, being effective against *S. aureus* but not against *S. epidermidis* (Fig. 11), and, displayed similar activity to C16:1Δ6 against *Propinibacterium acnes*, which is associated with AD (Ishibashi et al., 2009). In addition, 7-oxaheptadecanoic acid had gained an additional anti-yeast activity against *Malassezia*, which is associated with skin disorders such as seborrhea dermatitis and AD (Tajima et al., 2008). Moreover, the best molecule for treatment of skin disorders such as seborrhea dermatitis and AD could be selected by means of structure-activity relationships (Sugai et al., in preparation).

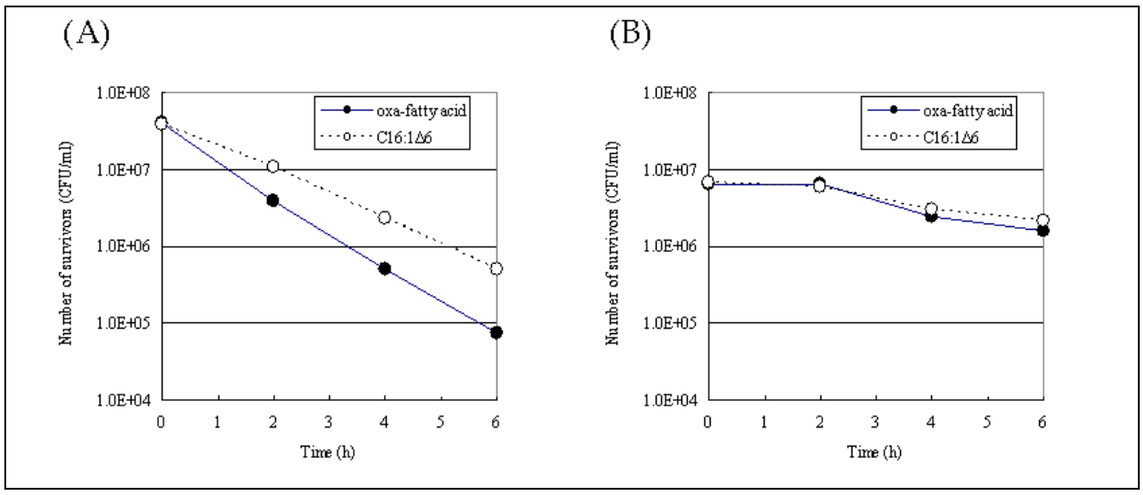

Fig. 11. Antibacterial activity of 7-oxaheptadecanoic acid. A: *S. aureus*; B: *S. epidermidis*

6. Conclusion

We found that *cis*-6-hexadecenoic acid (C16:1Δ6) displayed a selective antimicrobial activity that was unique among human sebaceous lipids. It was suggested that topical application of C16:1Δ6 to the skin of AD patients, who have lower levels of C16:1Δ6 in skin lipids than healthy controls, was effective for treatment of this skin condition. We proposed an industrial process for the production of C16:1Δ6 using *Rhodococcus* sp. An oxygen-containing analog of C16:1Δ6 (7-oxaheptadecanoic acid) was found to be more effective against microorganisms such as *S.aureus* and *Malassezia* sp. that are associated with skin disorders. It was proposed that the best oxa-fatty acid derivative of C16:1Δ6 could be selected by analysis of structure-activity relationships.

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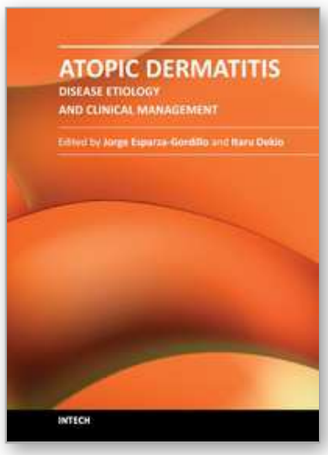

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Atopic Dermatitis is a common disease characterized by inflamed, itching and dry skin. This relapsing allergic disorder has complex etiology and shows a remarkably high clinical heterogeneity which complicates the diagnosis and clinical management. This book is divided into 4 sections. The first section (Disease Etiology) describes some of the physiological mechanisms underlying Atopic Dermatitis, including alterations in the immune system and the skin-barrier function. The important role of host-microorganism interactions on the pathophysiology of Atopic Dermatitis is discussed in the second section (Microorganisms in Atopic Dermatitis). An overview of the clinical diagnostic criteria and the disease management protocols commonly used is given in the third section (Diagnosis and Clinical Management). The last section (New Treatments) describes new therapeutic approaches that are not widely used but are currently being studied due to preliminary evidence showing a clinical benefit for Atopic Dermatitis.

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