UNITRIENOL T-27 AND T-272 WSL:

HAIR AND SKIN SEBUM NORMALIZER
UNITRIENOL

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UNITRIENOL

1. Introduction
Induchem Companies developed an oil soluble and a water soluble agent for the treatment of skin dryness and skin greasiness. The oil soluble Unitrienol T-27 is composed by Panthenyl Triacetate, Farnesol and Farnesyl Acetate. The water soluble Unitrienol T-272 WSL is composed by PEG-12 Glycerol Laurate, PEG-36 Castor Oil, Panthenyl Triacetate, Farnesol and Farnesyl Acetate. The two Unitrienol types are able to regulate the skin’s moisture and skin’s sebum content in cases of dry skin and oily skin. In fact, Panthenyl Triacetate, Farnesol, and Farnesyl Acetate were incorporated in Unitrienol formula due to their biological properties. Farnesol and Farnesol derivates are widespread in the plant world, in particular in the essential oil plant. Farnesol and Farnesol derivates are natural substances of high biological potency, which are also found in this form as bioactivators in human skin for squalene (appendix 1), and cholesterol formation (appendix 2). Panthenyl Triacetate and Farnesyl Acetate were respectively synthetized by acetylation of Panthenol and Farnesol. Panthenol and Panthenol Triacetate are converted in vivo into pantothenic acid[1]. Pantothenic acid is classed as part of the vitamin B complex and is a characteristic natural product of the human metabolism. Pantothenic acid is one of several precursor substances forming coenzyme A (appendix 3), which is an important cofactor for acylation reactions in numerous biochemical processes in the body and in particular, squalene (appendix 1) and cholesterol metabolism (appendix 2). The derivatives of Panthenol and Farnesol were used in the formula in order to provide a sustained action. Indeed, their conversion is more slowly in comparison to Panthenol and Farnesol into biological active molecules.

The present technical report contains the clinical assessments of Unitrienol T-27 as anti-skin dryness, anti-skin and anti-scalp/hair greasiness.

2. Oily skin/ oily scalp
The oily appearance of skin results from an excess of sebum excretion and spreading over the body surface and its interaction with the skin surface. Sebum is produced by sebaceous glands found throughout the human body except on the palms of the hands and soles of the feet. They are largest and most concentrated in the face and scalp where they are the sites of origin of acne and seborrhea. The normal function of sebaceous glands is to produce and excrete sebum, a group of complex oils including triglycerides and fatty acid breakdown products (major constituents about 57%), wax esters, squalene, cholesterol esters and cholesterol. Human sebum contains 13% of
squalene as one of its major constituents. Sebum lubricates the skin to protect against friction and makes it more impervious to moisture.

3. Dry skin
Dry skin, also known as xerosis cutis or asteatosis occurs worldwide and may affect patients of all ages, but it can probably be found in almost all patients over the age of 60. Dry skin results in a rough, scaly quality to the skin. Dry skin is due in part to various exogenous factors, such as dry climates, colder winter months, and excessive showering or bathing. Other factors, such as exposure to various alkali and detergents, particularly in patients with dispositional skin irritability, trigger or worsen the ability for skin to maintain moisture. When skin dryness is mild, it can be relatively asymptomatic, but when more pronounced, it may be associated with inflammation and superficial cracking causing unpleasant itching, stinging, and general discomfort[2]. Skin barrier function resides primarily within the top layer of the epidermis, also known as the stratum corneum or “horny layer.” While important for thermoregulation, gas exchange, and protection against pathogens, this external-most layer also serves to maintain proper hydration. In adults, deficient production of sebum and horny layer dysfunction contribute to the development of xerosis. Ghadially et al[3] found marked abnormalities in barrier integrity and barrier repair associated with dry skin in adults. Functional skin changes (altered ratios of fatty acids esterified to ceramide) were attributed to global deficiency in all key stratum corneum lipids and in particular ceramides.

4. Mode of action of Unitrienol
For greasy skin treatment:
Greasy skin is the consequence of an excess of sebum at the skin surface. This excess of sebum is due to its over excretion from sebocytes. The sebum is over excreted after the rupture of the weak sebocytes membrane. Weakness of cell follicle membrane occurred in skin pathologies as acne and dermatitis seborrheic. Cholesterol is part of all cell membranes, and one of its functions is to increase cell membrane stability and rigidity. Panthenol Triacetate, Farnesol and Farnesol Acetate included in Unitrienol induce a modulation of lipids metabolism and particularly cholesterol[1] [4, 5] and squalene metabolism [6].

The skin and scalp had a biotic commensal community (Staphylococcus spp., Propionibacterium spp., and Malassezia spp.)[7]. The presence of these microorganisms (malassezia restricta, fufur and globossa)[8] in important proportion on the greasy skin/ scalp provide an ideal medium (inflammatory, irritant) which lead
to the development of dandruff and/or seborrheic dermatitis or acne[9]. The sebum is degraded by oxygen and microorganisms found on the skin/scalp surface. Once degraded the sebum and skin surface lipids become cytotoxic and irritant due to free fatty acid liberation, thus provoking reactive follicular hyperkeratosis and epidermis alteration, the onsets of scalp and skin diseases [10, 11]. The skin and scalp microorganism spreading is limited by Unitrienol due to the intrinsic antifungal and antibacterial properties of the active ingredients Farnesol and Farnesol Acetate [12, 13] [14]. The anti-inflammatory property is provided by Panthenyl Triacetate[15, 16], Farnesol and its derivative [17] whereas the antioxidant properties are provided by Farnesol[18].

For dry skin treatment:
Dry skin is associated with altered barrier function and inflammation. Panthenyl Triacetate one of the components of Unitrienol is able to enhance skin barrier repair [15, 22] and decrease skin inflammation. Dry skin is also due to a deficient production of sebum and epidermis lipids. Panthenol Triacetate, Farnesol and Farnesol Acetate are all involved in lipids metabolism and precisely cholesterol, ceramides and squalene. By promoting squalene synthesis dry skin are hydrated. Indeed squalene is known to have also hydration properties[23].
# UNITRIENOL

## 5. Description of Unitrienol

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Unitrienol T-27</th>
<th>Unitrienol T-272 WSL</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Composition</strong></td>
<td>INCI name</td>
<td>CAS number</td>
</tr>
<tr>
<td></td>
<td>Farnesyl Acetate</td>
<td>29548-30-9</td>
</tr>
<tr>
<td></td>
<td>Farnesol</td>
<td>4602-84-0</td>
</tr>
<tr>
<td></td>
<td>Panthenyl</td>
<td>94089-18-6</td>
</tr>
<tr>
<td></td>
<td>Triacetate</td>
<td>94089-18-6</td>
</tr>
<tr>
<td></td>
<td>PEG-12 Glyceryl</td>
<td>59070-56-3</td>
</tr>
<tr>
<td><strong>Active content</strong></td>
<td>100%</td>
<td>33%</td>
</tr>
<tr>
<td><strong>Appearance</strong></td>
<td>Slightly viscous liquid</td>
<td></td>
</tr>
<tr>
<td><strong>Solubility</strong></td>
<td>Insoluble in water</td>
<td>Soluble in water</td>
</tr>
<tr>
<td></td>
<td>Soluble in ethanol, isopropyl</td>
<td>Soluble in ethanol, propylene glycol and glycerin</td>
</tr>
<tr>
<td></td>
<td>myristate, propylene glycol,</td>
<td></td>
</tr>
<tr>
<td></td>
<td>polyethylene glycol, Cetiol V, Miglyol</td>
<td></td>
</tr>
<tr>
<td></td>
<td>812 and vegetable oils.</td>
<td></td>
</tr>
<tr>
<td><strong>Possible formulations</strong></td>
<td>Emulsions (oil/water and water/oil), lotions, oils and sticks</td>
<td>Water based formula</td>
</tr>
<tr>
<td><strong>Dosage</strong></td>
<td>2-8%</td>
<td></td>
</tr>
<tr>
<td><strong>Storage</strong></td>
<td>Recommended storage temperature: 5-25°C</td>
<td></td>
</tr>
<tr>
<td><strong>Shelf life</strong></td>
<td>2 years</td>
<td></td>
</tr>
<tr>
<td><strong>Safety assessment</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>(Cf safety report for more details)</strong></td>
<td>Ocular irritation: BCOP model; Non irritant</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Skin irritation: Occlusive patch test; Non irritant</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Mutagenicity: Ames assay; Non mutagenic</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ready biodegradability: 83%</td>
<td></td>
</tr>
</tbody>
</table>
6. Clinical investigations of Unitrienol T-27

6.1. Introduction
Unitrienol T-27 was investigated on human volunteers in three studies for its hydration property and sebo-regulatory properties. The first study targeted the skin dryness by enhancement of skin moisture retaining capacity; the second study targeted skin dryness and skin oiliness by promoting sebum content normalization whereas the third study targeted the oily/hair scalp.

6.2. Targeting skin dryness: Enhancement of skin moisture retaining capacity

6.2.1. Materials and methods

6.2.1.1. Description of the panel and study condition
40 relatively elderly volunteers (20 for cream containing the active and 20 for placebo) were involved in the double-blinded study, half women and half men, largely exhibiting water-depleted skin. For three days before the investigation no skin preservation treatments were carried out on those part of the skin under investigation. During the measurements, care was taken to ensure that the volunteers were physically and mentally stable. Before and throughout the investigations the volunteers were not allowed to take tea or coffee or medicines.

A closed underground room was used as the investigation room. The room temperature during 5 hours measurement time was 20-22°C and the atmospheric humidity was 55-70%. The creams (containing active T-27 at 5% and placebo) were applied to 4 x 4 cm marked area on both left and right forearms and in amount of 0.5g/application and left for 1 min. The excess was then removed. Before application of the creams the moisture content of the untreated skin was measured. The measurement was started in every case at 8, beginning with the blank measurement and continued every half hour for 5 hours.

6.2.1.2. Hydration measurement test method
The measurements were carried out with Bingmer’s dermatometer in accordance with the method described by Winkler A and Wagener H.H[24]. The Tur-Buser skin moisture tester was used to measure the moisture level of epidermis. The measurement apparatus is based on the principle of the stray field capacitor. The skin moisture tester responds to the water content of the measurement object. The measurement apparatus is suitably calibrated before every measurements series with distilled water. In addition to the calibration the values obtained with the apparatus can
be read off immediately on a measurement range of 100 scale units. On this scale the measurement reading for pure water is 80 units and for dry medium it is 0 units. The water content of young skin is 10-12%, which correspond to a value of 70-80 scale divisions. An extremely dry skin containing about 6% water corresponds to a value of 40-55 scale divisions.

6.2.2. Results and discussion

6.2.2.1. Enhancement of skin moisture retaining capacity
The placebo cream produces a sharp drop in the moisture value after the initial application, as is the case with creams that do not contain any moisture retention additives. Unitrienol T-27 cream produced substantially longer moisture retention and higher moisture content in the epidermis during the same period. Unitrienol T-27 enables the skin to be hydrated twice longer than when the volunteers applied the placebo (Table 1 and fig 1).

<table>
<thead>
<tr>
<th>Time</th>
<th>Moisture content (mean ± standard deviation)</th>
<th>Unitrienol T-27 versus placebo</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Placebo</td>
<td>Unitrienol T-27</td>
</tr>
<tr>
<td>Before treatment (T0)</td>
<td>38.7 ± 5.3</td>
<td>39.6 ± 4.6</td>
</tr>
<tr>
<td>After 30 min</td>
<td>80 ± 0.3</td>
<td>80 ± 0.66</td>
</tr>
<tr>
<td>After 60 min</td>
<td>77.3 ± 1.38</td>
<td>79.6 ± 0.68</td>
</tr>
<tr>
<td>After 120 min</td>
<td>72.2 ± 1.7</td>
<td>78.2 ± 2.11</td>
</tr>
<tr>
<td>After 180 min</td>
<td>67.5 ± 1.9</td>
<td>75.2 ± 3.72</td>
</tr>
<tr>
<td>After 240 min</td>
<td>65.9 ± 1.7</td>
<td>73.1 ± 4</td>
</tr>
<tr>
<td>After 270 min</td>
<td>65.7 ± 2.5</td>
<td>70.9 ± 3.93</td>
</tr>
<tr>
<td>After 300 min</td>
<td>65.1 ± 2</td>
<td>68.9 ± 3.81</td>
</tr>
</tbody>
</table>

*According to Student “t” test p<0.05

Table 1: Moisture content of the skin after application of a cosmetic cream containing Unitrienol T-27

Figure 1: Moisture content of the skin after application of a cosmetic cream containing Unitrienol T-27
6.3. Targeting skin dryness and skin greasiness: sebum content normalization studies

6.3.1. Materials and methods

6.3.1.1. Description of the panel and study condition
Twenty subjects (10 males and 10 females) aged over 45 with dry skin and twenty subjects aged (10 males and 10 females) under 30 with predominantly seborrheic skin or mixed to greasy skin were included in the study. All subjects treated their skin with the cream containing the active ingredient at 5% twice a day during 45 days and then stopped its use in the 5 subsequent weeks. The skin greasy level was measured on the forehead about 2 cm above the nose. Individuals’ measurements were made at intervals of approximately 10 days, and no cream being used the day before the measurement and on the day itself.

6.3.1.2. Sebum investigation on human skin forehead (test method)
The quantity of sebaceous matter on the skin surface was measured using a “Creachem sebumeter” in accordance with the method developed by H. Schaefer and H. Kuhn-Bussius[25]. The principle of the method is based on the fact that when roughened glass or roughened plastic film is pressed against the skin it absorbs grease from the latter and thereby becomes transparent. The degree of transparency depends on the amount of grease on the skin and is measured photometrically. The sebumeter is in principle a grease spot photometer. By pressing a matte plastic strip against the skin some of the latter’s grease is taken up and can be photometrically measured by the brightening effect it produces on the roughened glass.

6.3.2. Results and discussion

6.3.2.1. Normalization of skin sebum content of dryness skin
When a cream containing Unitrienol T-27 at 5% is applied on a dry forehead, the content of sebaceous matter increased with time to achieve +12% (close to significance) after 30 days of treatment (table 2 and fig 2). Unitrienol T-27 must be used at least during 45 days to reveal a significant effect. The value obtained is the rate seen usually in normal skin. The efficiency of the treatment was seen in all subjects and seems to persist also after stopping the treatment.
6.3.2. Normalization of skin sebum content of greasy skin

When a cream containing Unitrienol T-27 at 5% is applied on a greasy forehead, the content of sebaceous matter is strongly reduced by about -36% after 30 days of treatment (table 3) achieving a rate seen usually in normal skin. The sebum reduction observed persists also after cessation of the treatment (fig 3). The efficiency of the treatment was seen in all subjects.

**Table 2: Content of sebaceous matter in the skin after application of a cosmetic cream containing the active ingredient and % of increase in comparison to day 0 (sebumeter analysis)**

<table>
<thead>
<tr>
<th>Time</th>
<th>Sebum quantity (arbitrary unit: mean ± standard deviation)</th>
<th>Average variation (%) vs D0</th>
<th>Student t test (D0 vs Dx)</th>
</tr>
</thead>
<tbody>
<tr>
<td>D0</td>
<td>17.85 ± 3.5</td>
<td>+2%</td>
<td>6,34E-01 non significant</td>
</tr>
<tr>
<td>After 10 days</td>
<td>18.2 ± 2.9</td>
<td>+4%</td>
<td>4,23E-01 non significant</td>
</tr>
<tr>
<td>After 20 days</td>
<td>18.95 ± 2.13</td>
<td>+7.5%</td>
<td>2,66E-01 non significant</td>
</tr>
<tr>
<td>After 30 days</td>
<td>19.2 ± 3.1</td>
<td>+12%</td>
<td>1,00E-01 close to significance</td>
</tr>
<tr>
<td>After 45 days</td>
<td>19.9 ± 4.33</td>
<td>+8.7%</td>
<td>2,00E-01 non significant</td>
</tr>
</tbody>
</table>

*According to Student "t" test p<0.1

**Table 3: Content of sebaceous matter in the skin after application of a cosmetic cream containing the active ingredient and % of decrease in comparison to day 0 (sebumeter analysis)**

<table>
<thead>
<tr>
<th>Time</th>
<th>Sebum quantity (arbitrary unit: mean ± standard deviation)</th>
<th>Average variation (%) vs D0</th>
<th>Student t test (D0 vs Dx)</th>
</tr>
</thead>
<tbody>
<tr>
<td>D0</td>
<td>28.87 ± 7.9</td>
<td>-26.00%</td>
<td>1,20E-04 Significant</td>
</tr>
<tr>
<td>After 10 days</td>
<td>21.3 ± 7.4</td>
<td>-39.00%</td>
<td>1,97E-07 Significant</td>
</tr>
<tr>
<td>After 20 days</td>
<td>17.4 ± 5.1</td>
<td>-36.00%</td>
<td>7,71E-10 Significant</td>
</tr>
<tr>
<td>After 30 days</td>
<td>18.36 ± 6.2</td>
<td>-36.8%</td>
<td>1,25E-07 Significant</td>
</tr>
<tr>
<td>After 45 days</td>
<td>18.26 ± 5.9</td>
<td>-44.8%</td>
<td>1,45E-05 Significant</td>
</tr>
</tbody>
</table>

*According to Student "t" test p<0.05
6.4. Targeting scalp greasiness: Anti-oily scalp study

6.4.1. Materials and methods

6.4.1.1. Description of the panel and study condition

28 healthy female and male adult volunteers with oily hair and scalp skin aged from 22 to 68 years old, with a mean sebum rate >80 µg/cm² were included in the study. Among 28 subjects, 14 used either shampoo with active ingredients at 2% or placebo during 28 days. Then they used their usual shampoo from D28 to D35. They were asked to use the shampoo 3 or 4 time per week on wet hair, to massage gently to lather and then rinsed off (estimated time of contact with the scalp: 5 min). Before each visit to the laboratory (D14, D28, D35), volunteers should wash their hair on the previous morning. The volunteers, using a questionnaire, assessed cosmetic performances of the shampoo.

6.4.1.2. Sebum investigation on human hair

Test method of assessment of the oily hair grade: The assessment of the efficacy of the shampoo was based on a clinical scoring of the "oily aspect of the hair roots" according a 7 grade scale (from R0 to R6). The grade R0 correspond to no oily aspect and the grade R6 correspond to very oily aspect. The scoring was performed at D0, D14, D28, and D35. The data were analyzed according the percentage of reduction of the clinical scoring retrieved at each study time. Quantitative sebumetric analysis were also performed on the forehead skin (median area at the hairline) and on the scalp of the volunteers with the Sebumeter SM 815® (Courage & Khazaka) at the beginning of the study (D0), after 14 days of use (D14) and after 28 days of use of the tested product (D28). Another measurement was taken on D35, 7 days after stopping the
treatment (use of their usual product). For each time (D0, D14, D28 and D35), 3 measurements were taken on the median forehead skin (median area at the hairline). The average and standard deviation of these three values were calculated and kept as the experimental value.

6.4.2. Results and discussion
When a shampoo containing Unitrienol T-27 at 2%, is applied on oily scalp skin, the content of sebaceous matter is strongly reduced (statistically significant) by about 40% after 28 days of treatment (table 4). Unitrienol T-27 showed an improvement of 8.3% in comparison to placebo treatment after 28 days of treatment. The sebum reduction observed persists also after cessation of the treatment (fig 4). The efficiency of the treatment was seen in quasi all subjects (13 out of 14).

<table>
<thead>
<tr>
<th>Time</th>
<th>Shampoo with active ingredient</th>
<th>Shampoo with placebo</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&quot;Clinical scoring of Oily aspect of the hair roots&quot; (mean ± standard deviation)</td>
<td>&quot;Clinical scoring of Oily aspect of the hair roots&quot; (mean ± standard deviation)</td>
</tr>
<tr>
<td></td>
<td>Average variation (%) vs D0</td>
<td>Student t test D0 vs Dx (p value and significance)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D0</td>
<td>4 ± 1</td>
<td>4 ± 0.96</td>
</tr>
<tr>
<td>After 14 days</td>
<td>3.31 ± 1.32</td>
<td>3.21 ± 0.97</td>
</tr>
<tr>
<td>After 28 days</td>
<td>2.38 ± 1.19</td>
<td>2.71 ± 0.79</td>
</tr>
<tr>
<td>After 35 days</td>
<td>2.23 ± 1.09</td>
<td>2.57 ± 1.09</td>
</tr>
</tbody>
</table>

Table 4: Clinical scoring of oily aspect of the hair roots after application of a shampoo containing the active ingredient or placebo and % of decrease in comparison to day 0.

![Image](image.png)
The self-assessment (fig 5) done by the volunteers confirmed the clinical scoring seen. The volunteers noticed the sebum reduction on their hair as well as a strong efficacy of the product, which purifies their scalps and hairs.

The efficiency of Unitrienol T-27 used as a lotion will probably give better results than those seen in this study using a shampoo formula. Indeed, the contact time of the active ingredient with the scalp is very short in the case of shampoo (less than 5 min and the product was rinsed) in comparison to lotion treatment known to be a leave on treatment without rinsing.

7. Conclusions
Unitrienol is a range of active ingredients (oil soluble for Unitrienol T-27 and water soluble for Unitrienol T-27 WSL) composed by a mixture of Panthenyl Triacetate (PTA), Farnesol and Farnesyl Acetate (FA). Panthenyl Triacetate and Farnesyl Acetate are converted in vivo into Panthenol and Farnesol. The derivatives of Panthenol and Farnesol were used in the formulas in order to provide a sustained action. Indeed, their conversion is more slowly in comparison to Panthenol and Farnesol into biological active molecules.

Clinical investigations performed on human volunteers having dry skin, greasy skin and greasy scalp have demonstrated the efficiency of Unitrienol (used at 2 and 5%) to treat each skin conditions.
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For dry skin condition Unitrienol:

• Improves 2 times the moisture retention capacity of the skin
• Normalize the content of sebaceous matter in the skin (+12%)
• The effect is persisting even 35 days after stopping the treatment

For greasy skin condition Unitrienol:

• Normalize the content of sebaceous matter in the skin (-36%)
• The effect is persisting even 35 days after stopping the treatment

For greasy scalp and hair condition Unitrienol:

• Decrease scalp sebaceous matter by 40% after 28 days of use (clinical scoring)
• The effect is persisting even 7 days after stopping the treatment (clinical scoring)
• Purifies the scalp and the hair (self assessment)
• Reduce the excess of sebum (self assessment)
• Had a degreasing effect (self assessment)
8. Appendix

Appendix 1

The starting material for sterol biosynthesis is acetyl coA.

Two molecules of acetyl coenzyme A condense to acetoacetyl coA.

![Diagram of acetyl coenzyme A condensation](image1)

The acetoacetyl-CoA condenses with a further molecule of acetyl-coenzyme A at the keto group, and 8-hydroxy-β-methylglutaric acid-coenzyme A is formed with the elimination of one molecule of coenzyme A.

![Diagram of 8-hydroxy-β-methylglutaric acid-coenzyme A formation](image2)

In a further step the CoA group is eliminated and the carboxyl group is converted to the alcohol, consuming 2 NADPH + 2 H⁺.

![Diagram of mevalonic acid formation](image3)

NADPH = reduced nicotinamide adenine dinucleotide phosphate
NADPH⁺ = nicotinamide adenine dinucleotide phosphate

The mevalonic acid formed is converted to active isoprene (isopentenyl pyrophosphate) via several stages. By the successive condensation of three active isoprenes, farnesyl pyrophosphate is formed.

![Diagram of active isoprene formation](image4)

The condensation of two molecules of farnesyl pyrophosphate produces squalene:

![Diagram of squalene production](image5)
Appendix 2

The squalene chain, folded in a particular way by intermediate stages, is transformed to lanosterol by hydroxylation and cyclization. Lanosterol is converted by the oxidative elimination of three methyl groups. Cholesterol forms the basis of all sterols:

![Figure 1: The formation of cholesterol](image)

Appendix 3

The biosynthesis of coenzyme A starting from panthotenic acid proceeds according the following scheme:

![Figure 1: Coenzyme A](image)
Figure 2: The biosynthesis of Coenzyme A[26]
UNITRIENOL

9. Bibliographic references


