

The influence of the test product on the key organisms of the respective body region was examined.

### Information about the tested product:

**Manufacturer:**

TRI-K Industries, Inc.  
2 Stewart Court  
NJ 07834 Denville  
USA

**Name of the product:**

Galguard Trident S

<b>Product type:</b>	Ingredient
<b>Application:</b>	Leave-on
<b>Dilution:</b>	1.2% in water and 3% glycerol; pH 5.5
<b>Sample received:</b>	23 March 2023
<b>Test Start:</b>	27 February 2023
<b>Test End:</b>	30 May 2023
<b>Test Standard:</b>	<b>MyMicrobiome Standard 18.11 Face / Body</b>
<b>Test result:</b>	<b>1.8</b>
<b>Certification:</b>	granted

## Test description

The MyMicrobiome Standard evaluates cosmetic and personal care products, that encounter the skin or mucous membrane, in terms of their influence on the microbiome located at a specific body site.

An intact skin microbiome has a fundamental influence on skin health. Products which are to be skin-friendly must also be Microbiome-friendly in order not to unbalance the skin of the user.

The MyMicrobiome Standard evaluates the influence of cosmetic and personal care products on the microbial key players of a specific skin or mucous membrane area. The human microbiome is very individual from person to person.

Each area, however, harbors a characteristic composition of bacteria, viruses and fungi. The test examines the products influence on the key organisms typical for each skin area and thus offers a standardized procedure.

## Various aspects are examined:

### The microbial quality of the product.

#### The influence of the product on the natural, healthy skin balance.

The skin-commensal bacterium *Staphylococcus epidermidis* keeps the skin with antimicrobial peptides (so-called bacteriocins) and pH adjustments healthy and keeps skin-harmful germs such as *Staphylococcus aureus* in check. The product should not disturb this balance between skin-friendly and skin-harmful bacteria. This sensitive balance is investigated in conjunction with the product.

#### The influence of the product on the bacterial diversity of the specific body region.

Each body region is colonized by a certain microbial composition. For a healthy skin it is particularly important to maintain this biodiversity. The influence of the product on the respective microbial mixture is examined in the test. The aim is to find as many key organisms as possible after contact with the product.

#### The influence of the product on the growth behavior of the microbes of the specific body region.

In addition to the diversity of the specific microbiome, the growth or number of different key organisms should not be influenced by the product. This is investigated in a skin-product contact model. The key organisms are brought into direct and indirect contact with the product and their growth is observed.

### Results

#### The microbial quality of the product.

The prerequisite for the test for microbial friendliness is the microbiological quality of the product. The following table contains the limit values that must be observed.

Types of organisms	Limit values	
	Products specially designed for children under 3 years, eye area or mucous skins	Other products
<b>Total counts mesophilic, aerobic microorganisms (bacteria, yeasts, molds, (TAMC and TYMC))</b>	$\leq 1 \times 10^2$ cfu/g or ml <sup>a</sup>	$\leq 1 \times 10^3$ cfu/g or ml <sup>b</sup>
<i>Escherichia coli</i>	Not detectable in 1g or 1 ml	Not detectable in 1g or 1 ml
<i>Pseudomonas aeruginosa</i>	Not detectable in 1g or 1 ml	Not detectable in 1g or 1 ml
<i>Staphylococcus aureus</i>	Not detectable in 1g or 1 ml	Not detectable in 1g or 1 ml
<i>Candida albicans</i>	Not detectable in 1g or 1 ml	Not detectable in 1g or 1 ml
<b>a &gt;200 cfu/g or ml, b &gt;2000 cfu/g or ml</b>		

#### Results Microbiological quality

Determination of TAMC, TYMC, absence of *E. coli*, *P. aeruginosa* and *S. aureus*.

**The microbiological quality of the product according to DIN EN ISO 17516 is fulfilled.**

Parameter	Sample no.: 23.583.18.4
TAMC [cfu/0,1 ml]	< 1,0E+01
TYMC (incl. <i>Candida albicans</i> ) [in 0,1 ml]	negative
<i>Escherichia coli</i> [in 0,1 ml]	negative
<i>Pseudomonas aeruginosa</i> [in 0,1 ml]	negative
<i>Staphylococcus aureus</i> [in 0,1 ml]	negative

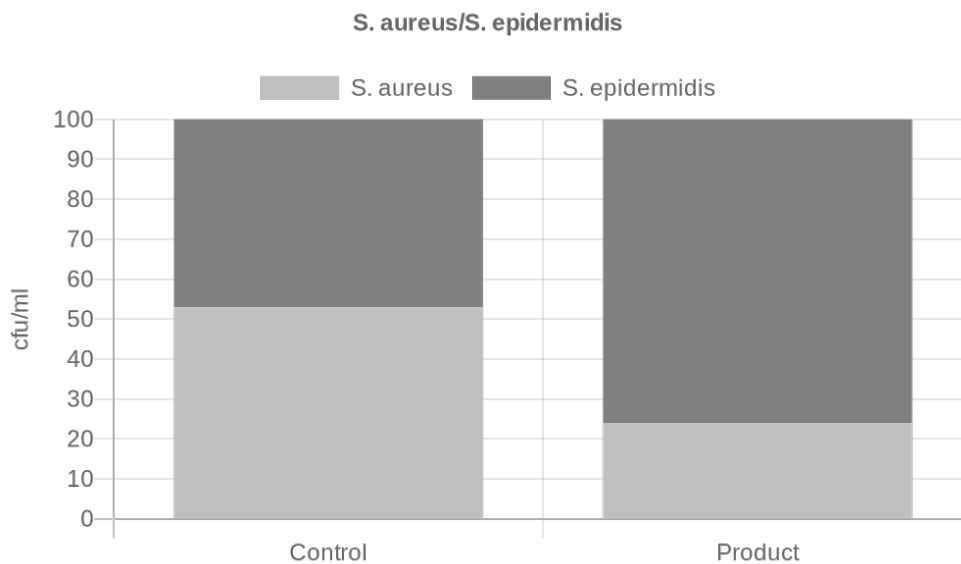
## Results

### The influence of the product on the natural, healthy skin balance.

A co-culture of *S. epidermidis* and *S. aureus* is incubated with the product.

The ratio of the two microbes to each other is determined.

Determination of the bacterial count at time t = 15 min (rinse-off) or 4h (leave-on).

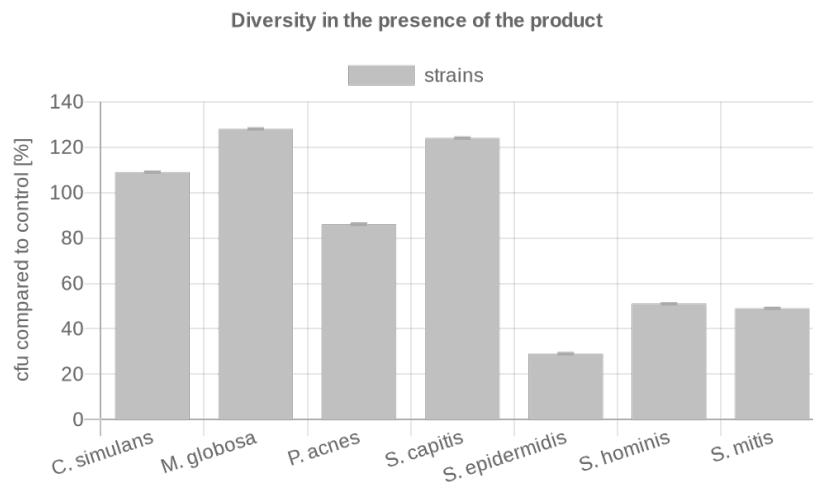


	cfu/ml		Ratio Product/ Control	Grade
	<i>S. aureus</i>	<i>S. epidermidis</i>		
<b>Control</b>	28933.3	25700	3.5	<b>1</b>
<b>Product</b>	850	2655		

## Results – SEBACEOUS SKIN -

### The influence of the product on the microbial diversity of the specific body region.

A co-culture of key organisms of the specific body region is incubated with the product for t = 15 min (rinse-off) or 4h (leave-on). The ratio of the microbes compared to the control (PBS) is determined.

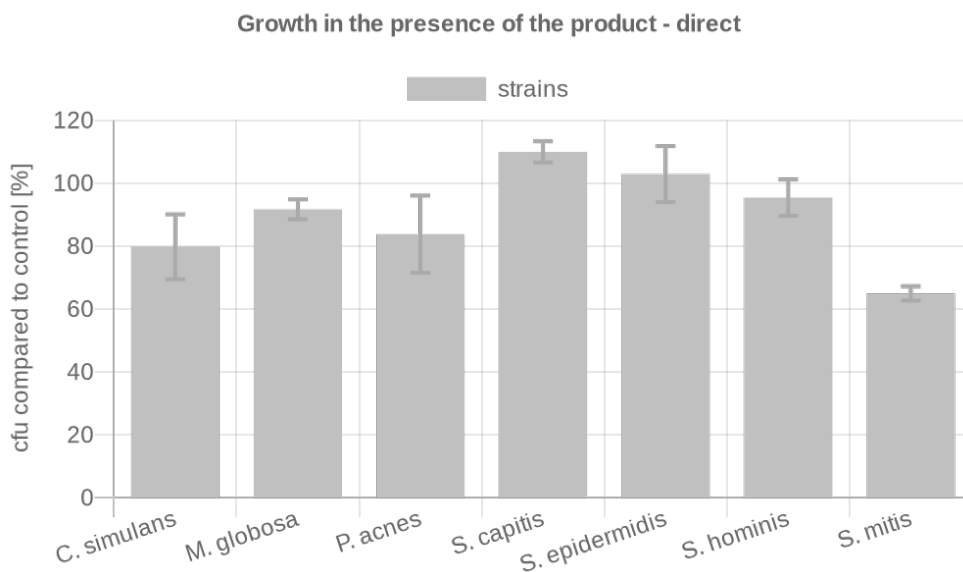


Key-Microbe	t=	4h	Rating
	cfu/ml		
<i>C. simulans</i>	Control	1433.3	1
	Product	1566.7	
<i>M. globosa</i>	Control	4360	2
	Product	5580	
<i>P. acnes</i>	Control	525	2
	Product	453.3	
<i>S. capitis</i>	Control	1166.7	1
	Product	1450	
<i>S. epidermidis</i>	Control	3766.7	3
	Product	1100	
<i>S. hominis</i>	Control	2266.7	3
	Product	1150	
<i>S. mitis</i>	Control	6.7	3
	Product	3.3	
<b>Overall rating:</b>			<b>2.1</b>

### Results – SEBACEOUS SKIN -

#### The influence of the product on the growth behavior of the microbes of the specific body region – directly.

The influence of the product on the growth of each individual microbe of the key organisms of the specific body region is investigated and put in relation to the control (PBS). Product contact with the microorganisms is directly.

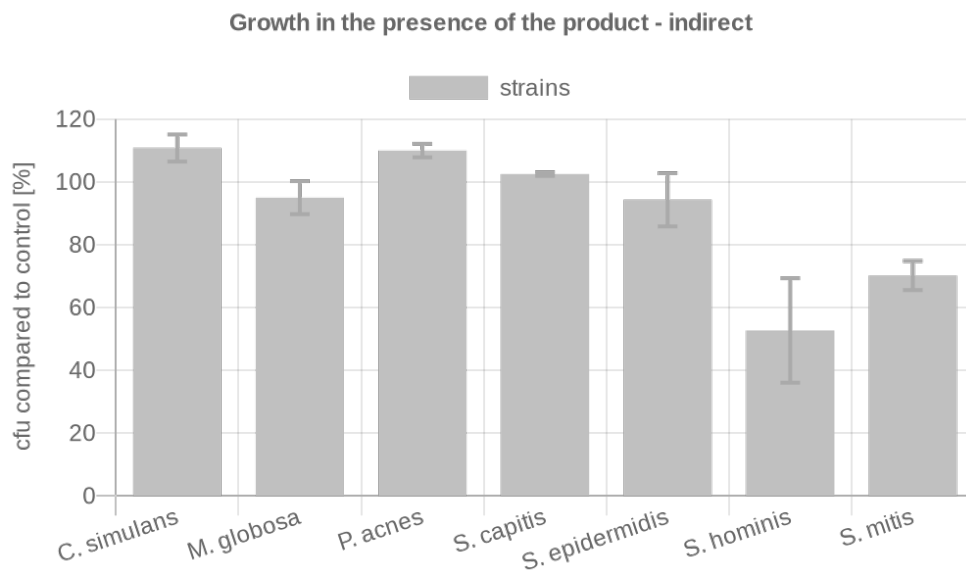


Key-Microbe	cfu/ml		Rating
	Control	Product	
<i>C. simulans</i>	Control	189.3	2
	Product	151	
<i>M. globosa</i>	Control	100	2
	Product	91.7	
<i>P. acnes</i>	Control	315.7	2
	Product	264.7	
<i>S. capitis</i>	Control	122.7	1
	Product	135	
<i>S. epidermidis</i>	Control	251.3	1
	Product	258.7	
<i>S. hominis</i>	Control	325.3	1
	Product	310.3	
<i>S. mitis</i>	Control	330	2
	Product	214.5	
<b>Overall rating:</b>			<b>1.6</b>

## Results – SEBACEOUS SKIN -

### The influence of the product on the growth behavior of the microbes of the specific body region – indirectly.

The influence of the product on the growth of each individual microbe of the key organisms of the specific body region is investigated and put in relation to the control (PBS). The product contact to the microorganisms is indirect.

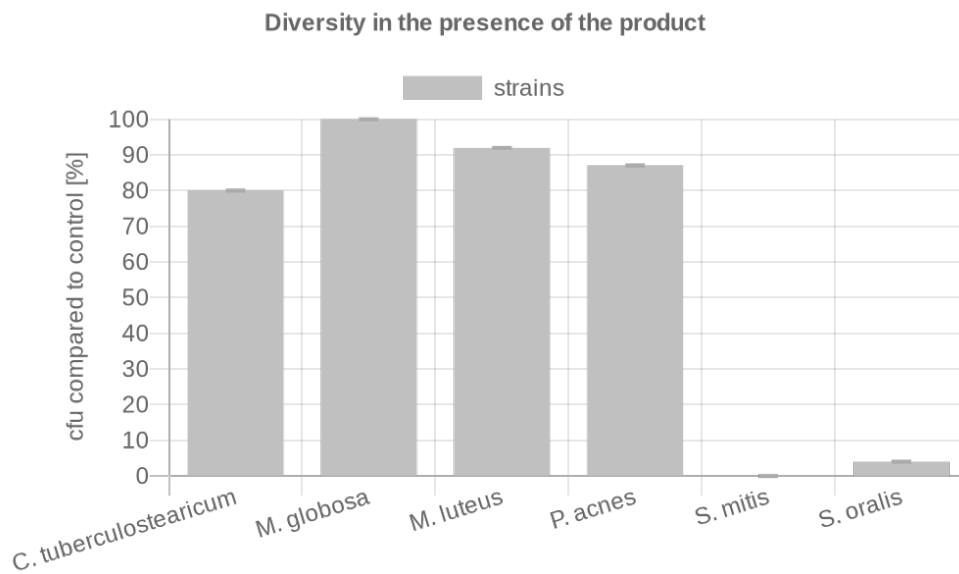


Key-Microbe	cfu/ml		Rating
<i>C. simulans</i>	Control	134	1
	Product	148.5	
<i>M. globosa</i>	Control	100	1
	Product	95	
<i>P. acnes</i>	Control	185.5	1
	Product	204	
<i>S. capitis</i>	Control	120.5	1
	Product	123.5	
<i>S. epidermidis</i>	Control	249.3	2
	Product	235	
<i>S. hominis</i>	Control	315	3
	Product	166	
<i>S. mitis</i>	Control	264.5	2
	Product	185.7	
<b>Overall rating:</b>			<b>1.6</b>

## Results – DRY SKIN -

### The influence of the product on the microbial diversity of the specific body region.

A co-culture of key organisms of the specific body region is incubated with the product for t = 15 min (rinse-off) or 4h (leave-on). The ratio of the microbes compared to the control (PBS) is determined.



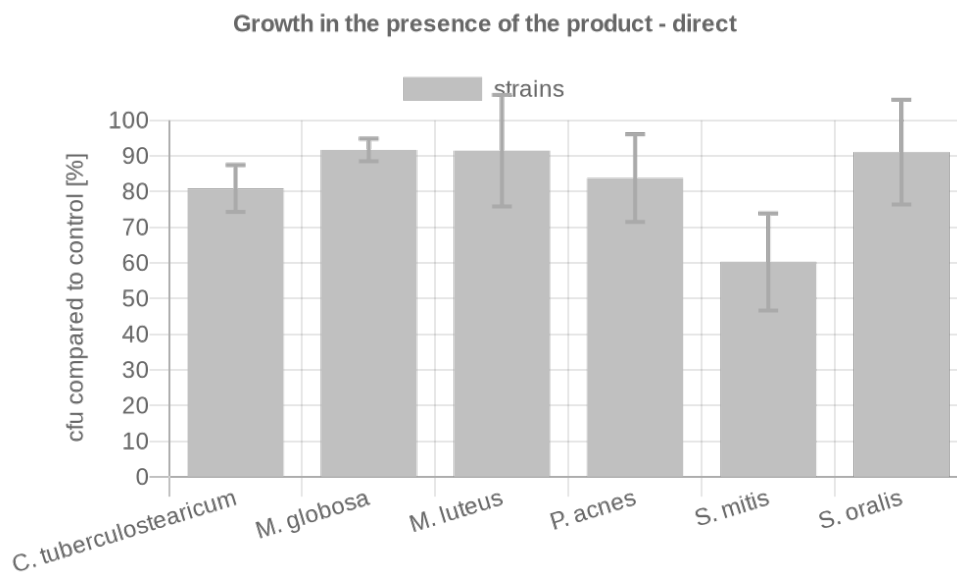
Key-Microbe	t=	4h	Rating
	cfu/ml		
<b>C. tuberculostearicum</b>	Control	683,3	2
	Product	550	
<b>M. globosa</b>	Control	1000	1
	Product	1000	
<b>M. luteus</b>	Control	1625	2
	Product	1500	
<b>P. acnes</b>	Control	663,3	2
	Product	576,7	
<b>S. mitis</b>	Control	n/a	n/a
	Product	n/a	
<b>S. oralis</b>	Control	470	3
	Product	20	
<b>Overall rating:</b>			<b>2</b>



### Results – DRY SKIN -

#### The influence of the product on the growth behavior of the microbes of the specific body region – directly.

The influence of the product on the growth of each individual microbe of the key organisms of the specific body region is investigated and put in relation to the control (PBS). Product contact with the microorganisms is directly.

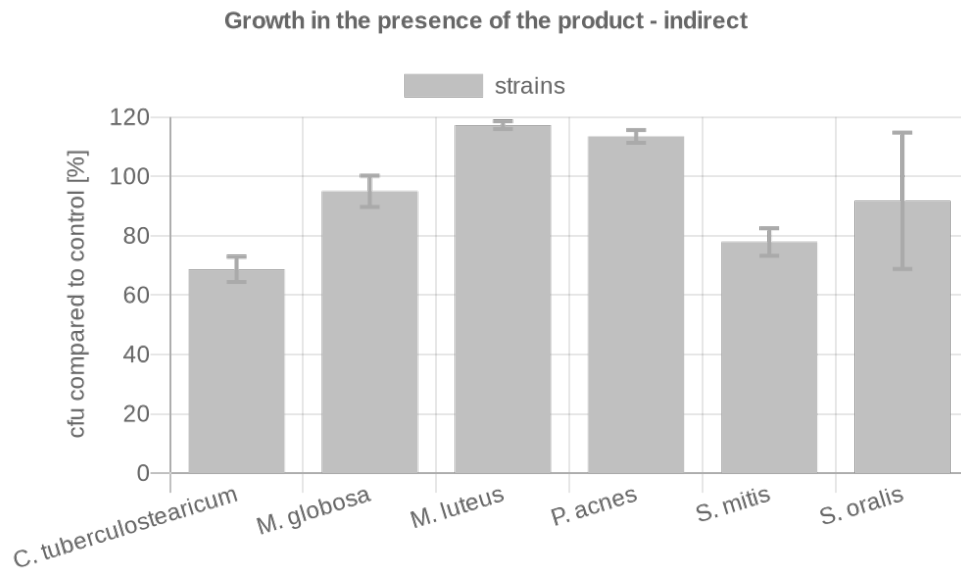


Key-Microbe	cfu/ml		Rating
	Control	Product	
<b><i>C. tuberculostearicum</i></b>	Control	1761	2
	Product	1424	
<b><i>M. globosa</i></b>	Control	100	2
	Product	91.7	
<b><i>M. luteus</i></b>	Control	549.3	2
	Product	502.7	
<b><i>P. acnes</i></b>	Control	315.7	2
	Product	264.7	
<b><i>S. mitis</i></b>	Control	330	3
	Product	199	
<b><i>S. oralis</i></b>	Control	15.7	1
	Product	14.3	
<b>Overall rating:</b>			<b>2</b>

## Results – DRY SKIN -

### The influence of the product on the growth behavior of the microbes of the specific body region – indirectly.

The influence of the product on the growth of each individual microbe of the key organisms of the specific body region is investigated and put in relation to the control (PBS). The product contact to the microorganisms is indirect.



Key-Microbe	cfu/ml		Rating
<i>C. tuberculoostearicum</i>	Control	1648.5	2
	Product	1133.3	
<i>M. globosa</i>	Control	100	1
	Product	95	
<i>M. luteus</i>	Control	137.7	1
	Product	161.5	
<i>P. acnes</i>	Control	179.7	1
	Product	204	
<i>S. mitis</i>	Control	238.3	2
	Product	185.7	
<i>S. oralis</i>	Control	40.3	1
	Product	37	
<b>Overall rating:</b>			<b>1.3</b>

### Results

The results are evaluated with grades from 1 (one) to 3 (three). If the product shows no or positive influence to the above-mentioned aspects, a grade of 1 is awarded respectively.

If only a very weak negative influence can be detected in the tests, the grade 2 is awarded and in case of a clearly negative influence, the product receives the grade 3.

The product has passed up to grade 2.0.

Here the grade means

1.0 – 2.0 = Microbiome-friendly; 2.1 – 3.0 = Microbiome-damaging.

Test	Grade
Balance of the skin microbiome	1
Diversity of the corresponding skin microbiome (sebaceous, x2)	2.1
Diversity of the corresponding skin microbiome (dry, x2)	2
Skin-product contact direct (sebaceous, x2)	1.6
Skin-product contact direct (dry, x2)	2
Skin-product contact indirect (sebaceous)	1.6
Skin-product contact indirect (dry)	1.3
<b>Overall grade</b>	<b>1.8</b>

**With an overall grade of 1.8 the seal „Microbiome-friendly“ is awarded according to MyMicrobiome Standard 18.11 Face / Body.**

Place, Date: Balzers, 30 Mai 2023

Responsible person: Dr. Kristin Neumann

Signature:

