

Using Next generation DNA Sequencer, this study shows the before and after results by skin flora analysis, from treating Acne with Nitrogen Plasma.

AgeJet

Accurate Nitrogen Plasma delivery

Method

- Nitrogen Plasma was used on the cheeks of 10 subjects.

The Skin was irradiated with 1Joule. Air Off.

- After an initial test taken prior to treatment, further test were taken after 24 hours and 72 hours. Indigenous bacteria were collected from an area $\phi 3$ cm at a distance of 1 cm from the lower eyelid and side of the nose, using dedicated scale template.

Method

Fungal flora analysis

16S rDNA after fragmenting and amplifying a DNA sample rRNA V2, 3, 4, 5, 6, 7, 8, 9 region sequence

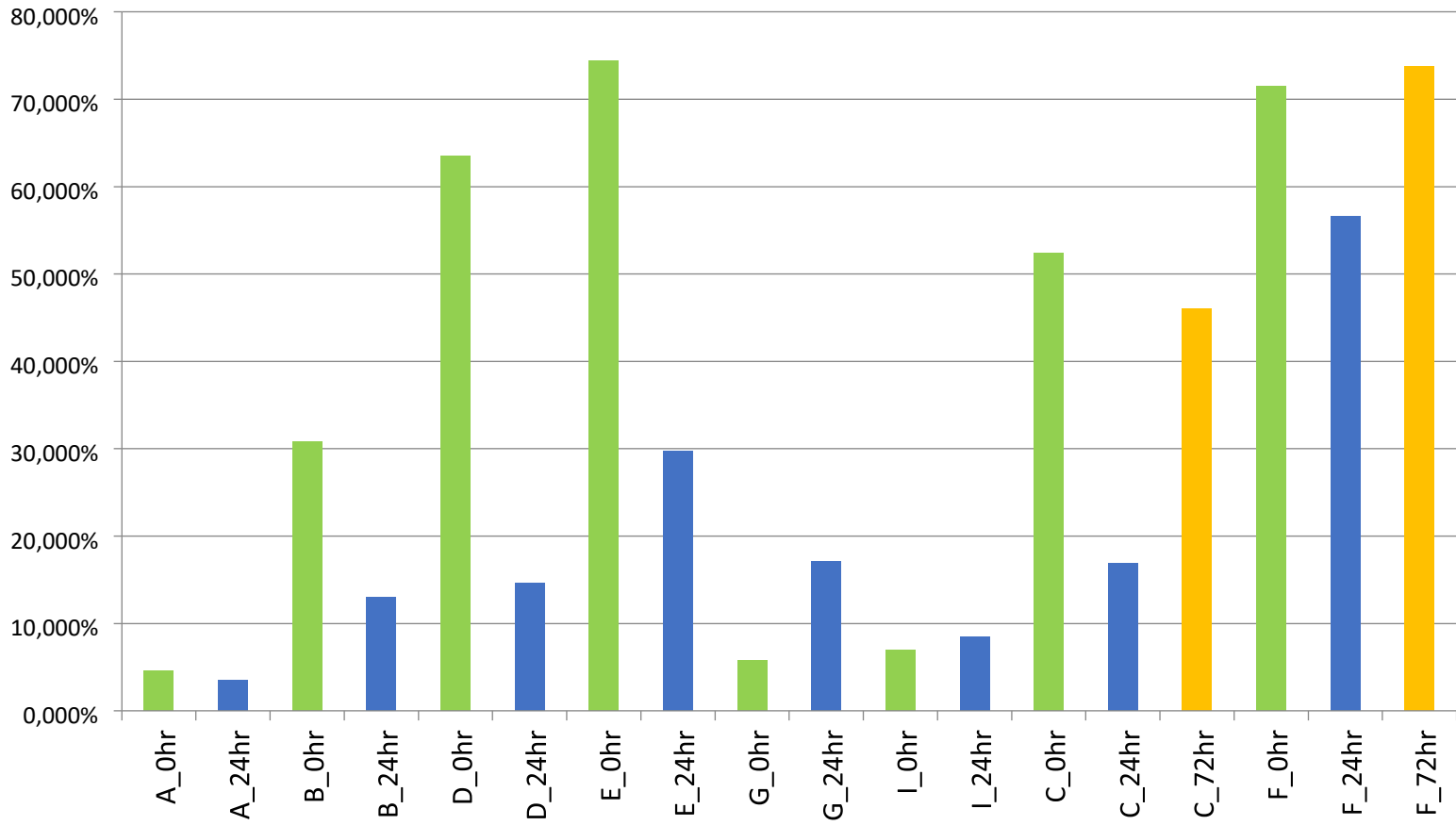
ION PGM Read the number of reads with the sequencer

Low quality base, read length 120bp or less were removed from the total bacterial count analysis.

A Takara bacteria Quantitative PCR kit was used to determine by the Real time PCR method.

Provided by: Chiharu Dermatology Clinic, Dr. Chiharu Watanabe

Propionibacterium acnes



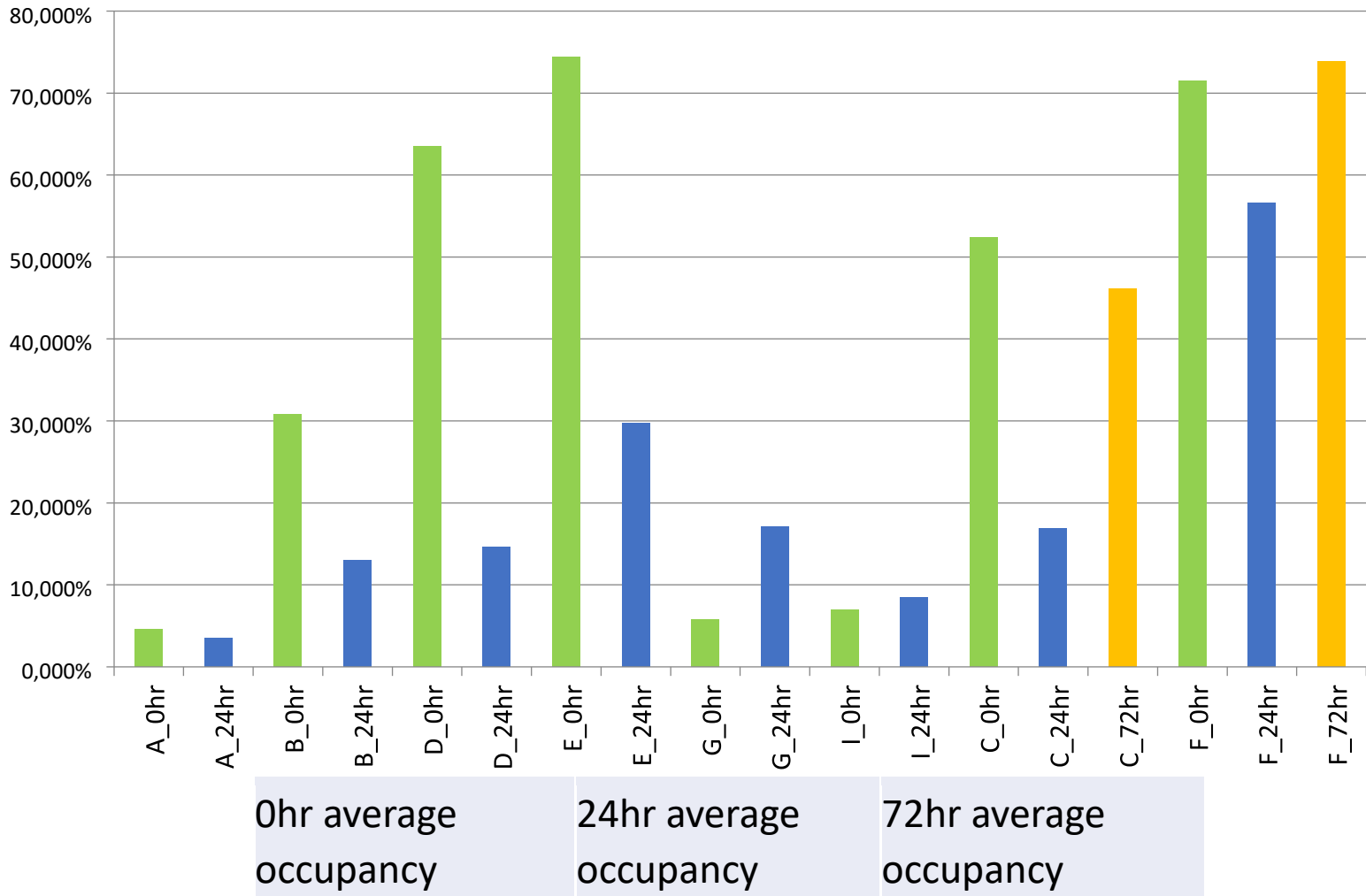
Results-Time series changes of major bacterial species-

Comparison between 0h and 24h: $P = 0.054$, shows a decrease in *P. acnes** Swab sample errors excluded (H, J subjects), presumed to be caused by subject collection method and DNA deterioration (fragmentation)

Provided by: Chiharu Dermatology Clinic, Dr. Chiharu Watanabe

Results- Show time series changes of major bacterial species-

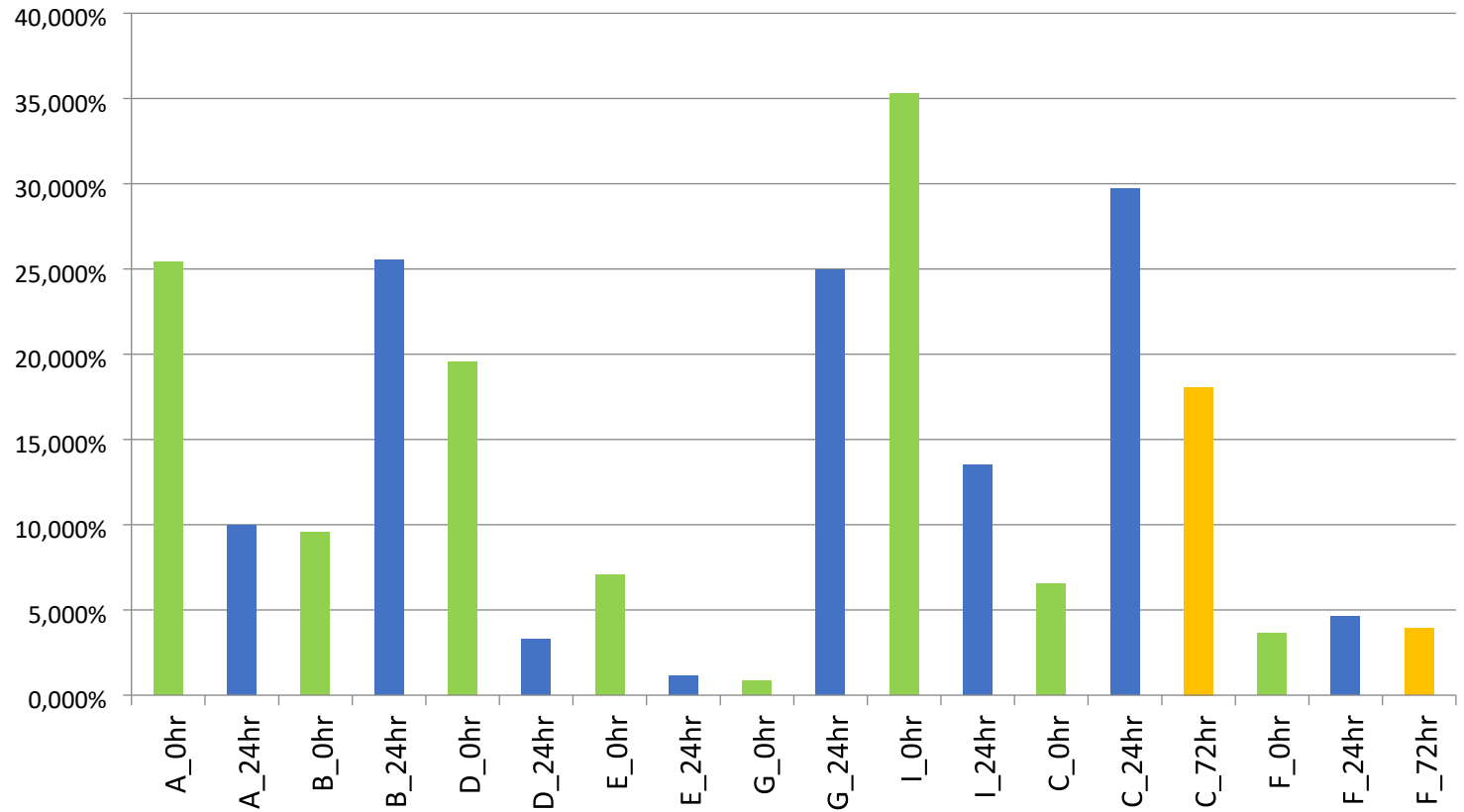
Propionibacterium acnes



Results-Time series changes of major bacterial species-



Chiharu Dermatology Clinic, Dr. Chiharu Watanabe



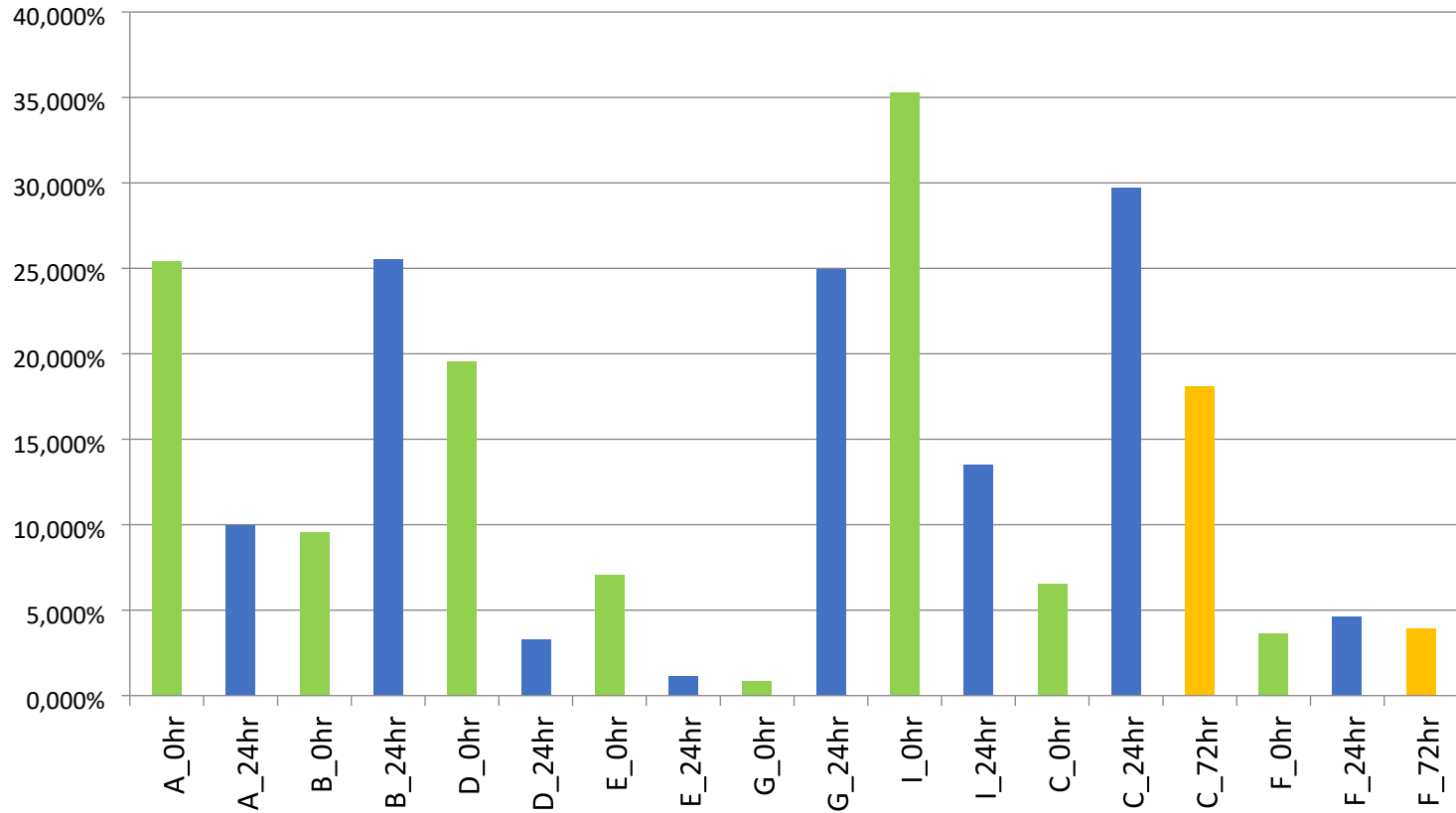
Staphylococcus epidermidis

Staphylococcus epidermidis, which is said to be a good bacterium, does not show a decreasing tendency (P = 0.87).)

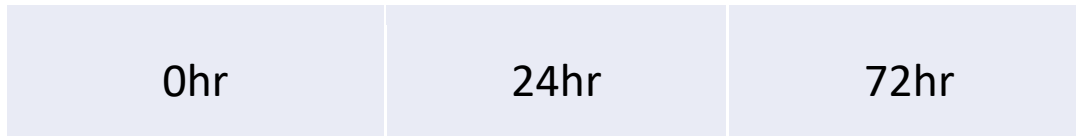
Results-Time series changes of major bacterial species-

Chiharu Dermatology Clinic, Dr. Chiharu Watanabe

Results-Time series changes of major bacterial species-



Staphylococcus epidermidis



Results-Time series changes of major bacterial species-

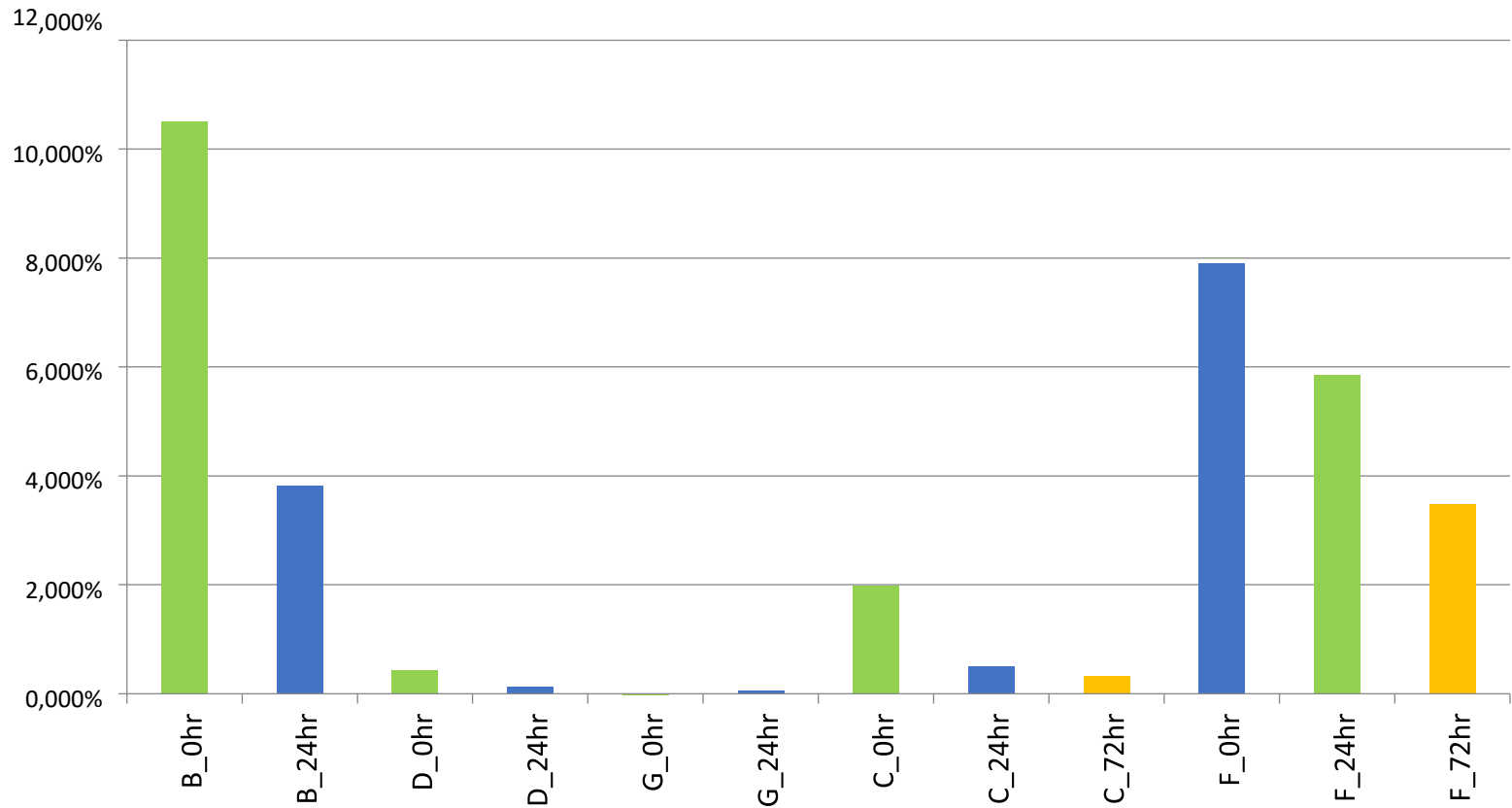
10.8%

11.3%

10.1%

Chiharu Dermatology Clinic, Dr. Chiharu Watanabe

Staphylococcus aureus



0hr	24hr	72hr
2.528%	1.413%	0.986%

* Results of only 5 atopic patients because Staphylococcus aureus is rarely detected in healthy subjects

Chiharu Dermatology Clinic, Dr. Chiharu Watanabe

Bacteria Name	0hr average diversity index	24h average diversity index	P value 0hr vs 24hr (Paired T-Test)
Staphylococcus epidermidis	1769	1452	0.828
Propionibacterium acnes	6271	1274	0.25
Staphylococcus aureus	544	200	0.352
Streptococcus thermophilus	6	1	0.352

Results-Time series changes of major bacterial species-

24 hours after plasma irradiation, all four bacterial species decreased, but P. acnes and Staphylococcus aureus decreased significantly.

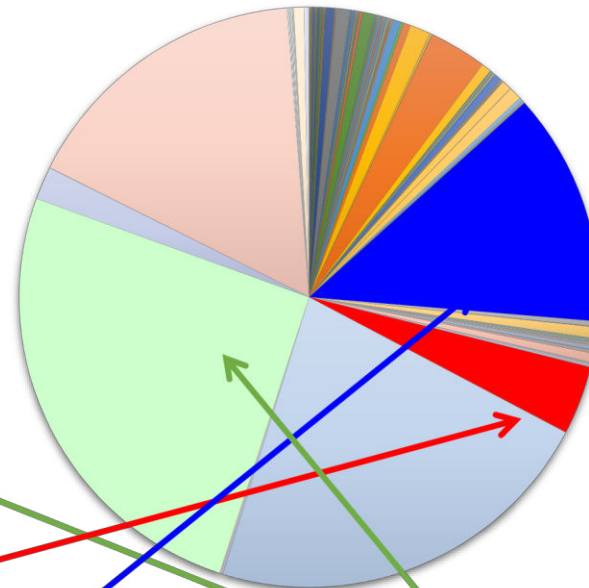
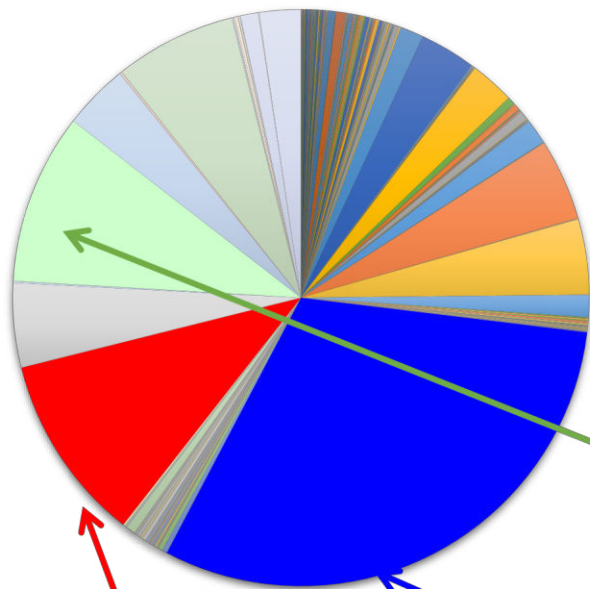
Sample Name	0hr average diversity index	24hr average diversity index	P value 0hr vs 24hr (Paired T-Test)
Simpson's Index	0.734	0.845	0.045

24 hours after plasma irradiation, the diversity index of indigenous skin flora increased. The balance is improved

Time-series changes of all strains pie
data, Atopic Dermatitis patient
Patient B

0時間

24時間



- Propionibacterium acnes
- Staphylococcus epidermidis
- Staphylococcus capitis
- Staphylococcus pasteurii

Staphylococcus aureus
11%→4%

Propionibacterium
31%→13%

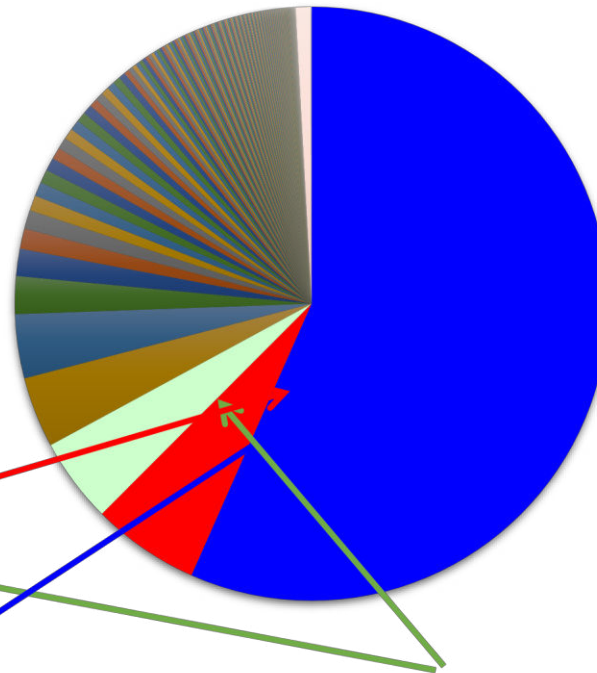
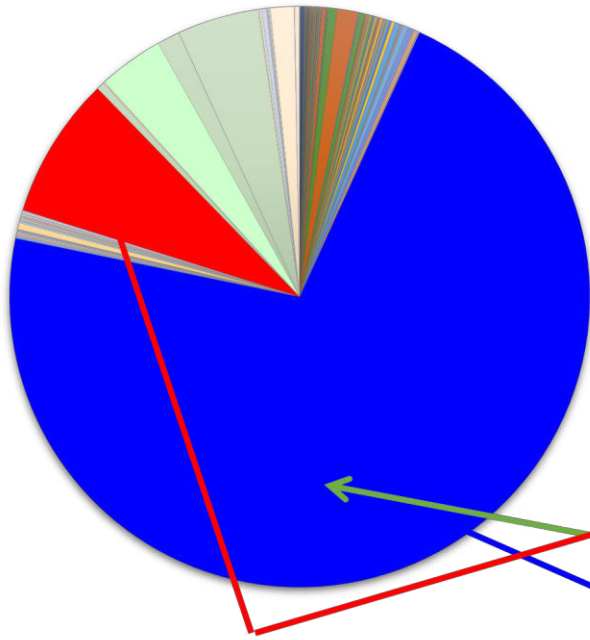
Staphylococcus epidermidis
10%→26%

acnes

Patient F

0時間

24時間



- Propionibacterium acnes
- Staphylococcus epidermidis
- Staphylococcus aureus
- Staphylococcus pasteurii

Staphylococcus aureus
8%→6%

Propionibacterium
71%→57%

Staphylococcus epidermidis
4%→5%

acnes

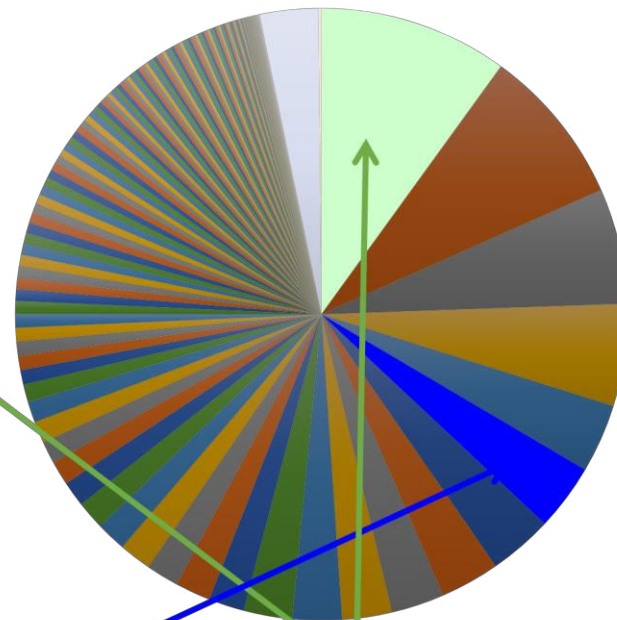
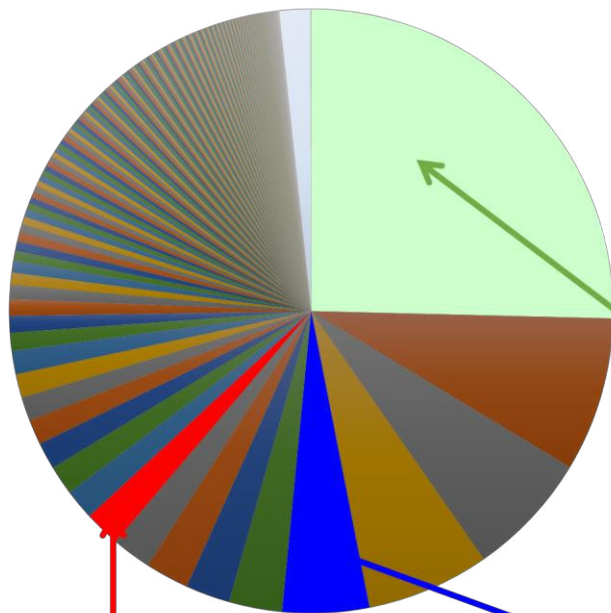
Chiharu Dermatology Clinic, Dr. Chiharu Watanabe

Time-series changes of all strains pie data ACNE patients

Patient A

0時間

24時間



- Staphylococcus emidermidis
- Staphylococcus pasteurii
- Staphylococcus hominis
- Corynebacterium tuberculostearicum

Staphylococcus aureus
2%→0.4%

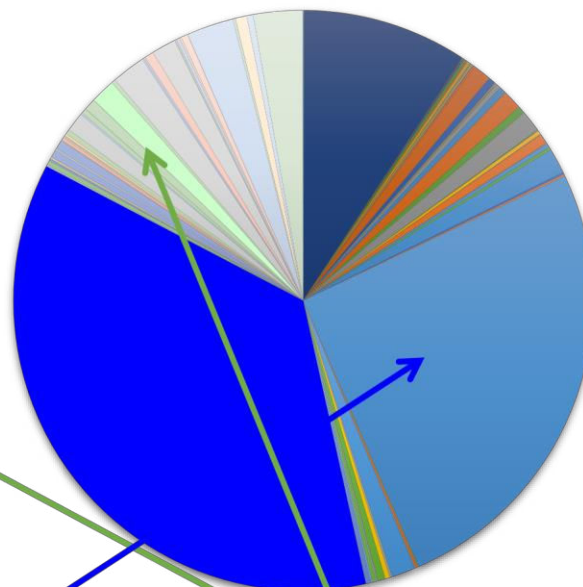
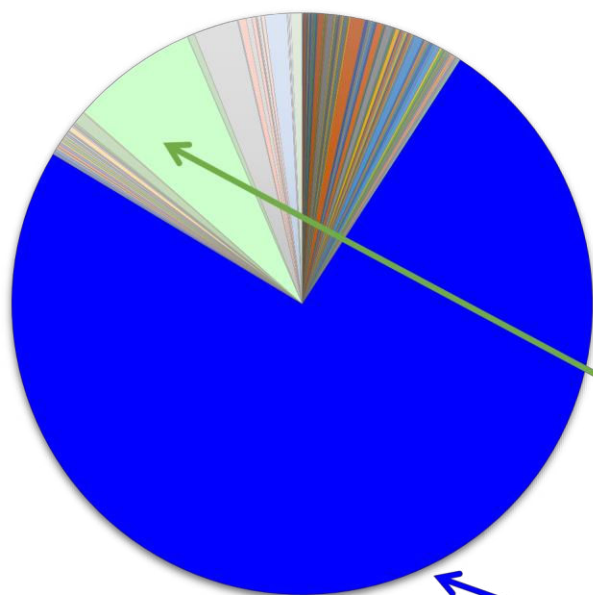
Propionibacterium acnes
5%→4%

Staphylococcus emidermidis
25%→10%

Patient E

0時間

24時間



- Propionibacterium acnes
- Staphylococcus epidermidis
- Lactobacillus nagelii
- Acetobacter fabarum

Staphylococcus aureus
0%→0%

acnes

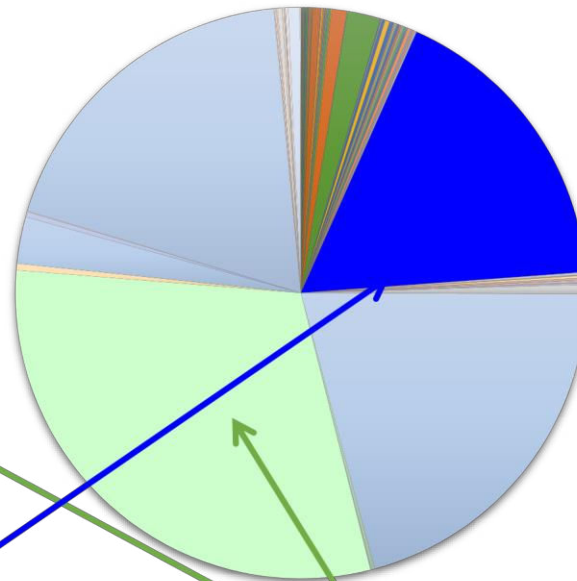
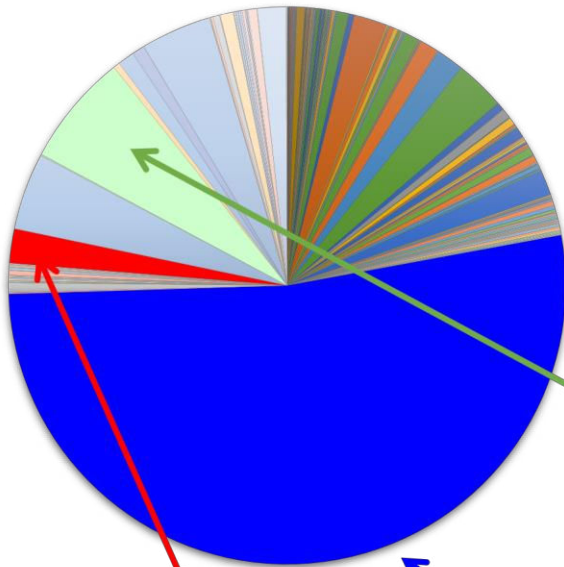
Propionibacterium
74%→36%

Staphylococcus epidermidis
7%→1%

Patient C

0時間

24時間



Staphylococcus aureus
2%→0.5%

Propionibacterium acnes
52%→17%

Staphylococcus emidermidis
7%→30%

Propionibacterium acnes

Staphylococcus epidermidis

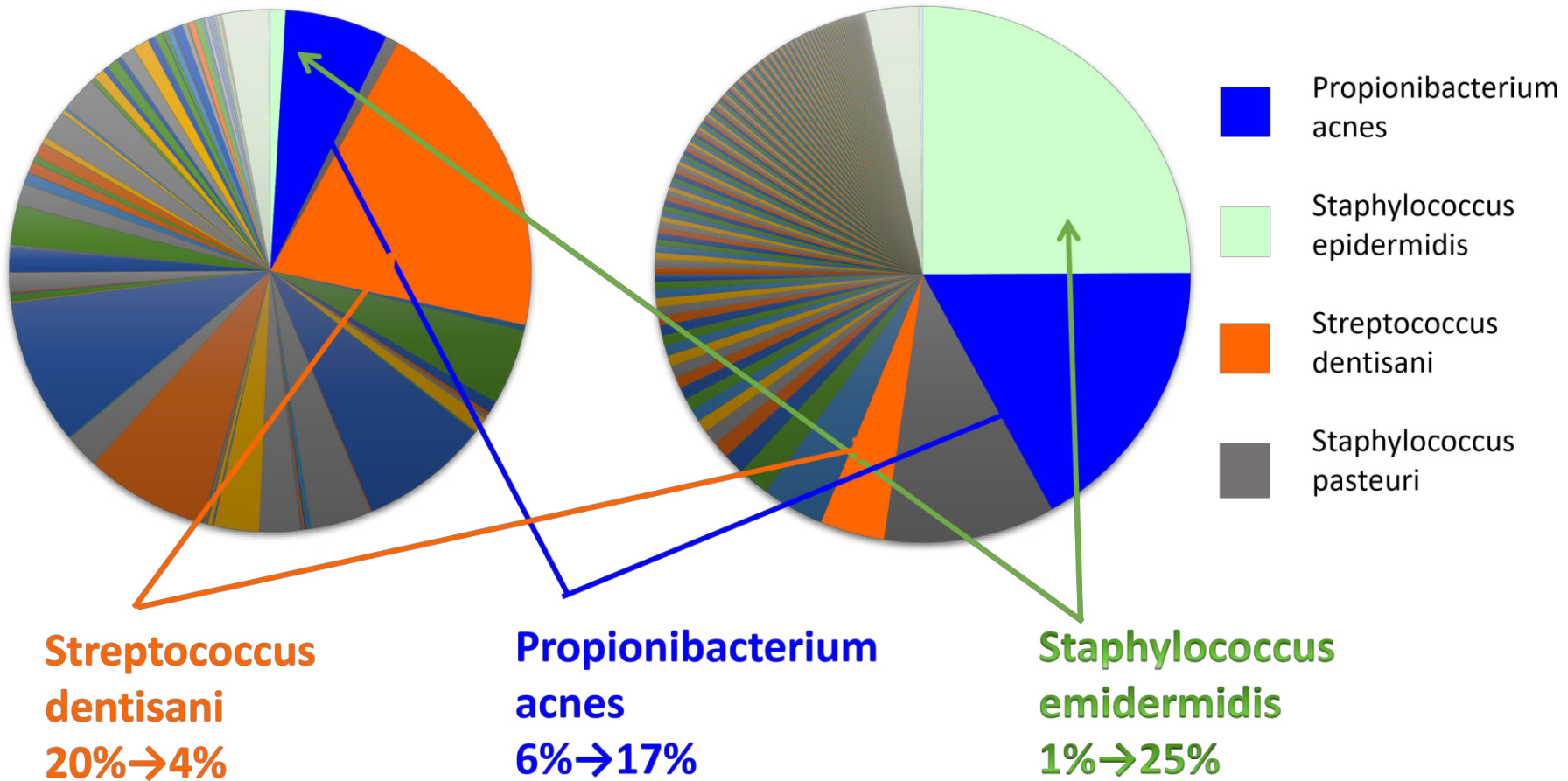
Staphylococcus aureus

Staphylococcus pasteurii

Patient G

0 時間

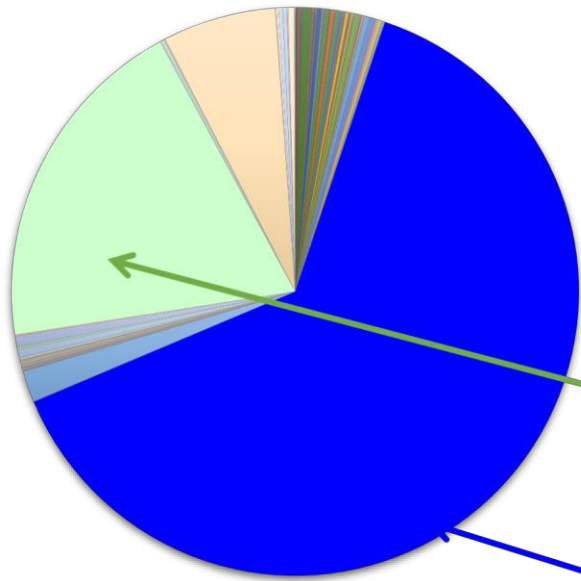
24 時間



Patient D

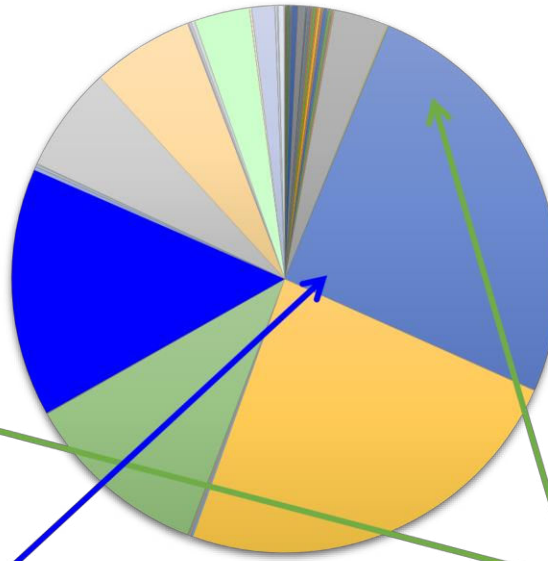
0 時間

24 時間



Staphylococcus aureus
0%→0%

Propionibacterium acnes
64%→15%



Staphylococcus emidermidis
20%→3%

- Propionibacterium acnes
- Staphylococcus epidermidis
- Pantoea vegans
- Staphylococcus pasteurii

Other / considerations

-From the subjects who performed skin flora analysis, it can be said that *P. acnes* and *Staphylococcus aureus* may be reduced by nitrogen gas plasma treatment, leading to a normal balance of indigenous skin bacteria. (However, it may reduce *Staphylococcus epidermidis*, so be careful about postoperative dryness.)

(Interesting case: G subject) • Group A haemolytic streptococcus, which is a reddish poison, is present in about 25% at 0 hours, and *Staphylococcus epidermidis* is very few, but after 24 hours, streptococcus is reduced to just over 5%, and staphylococcus is 40. It has increased to just over%. Streptococcus was also found to be sterilizable.

There is a possibility that the number of *P. acnes* will partially return after 72 hours. This time it was 0.8J / cm treatment, but the decreasing trend is maintained by increasing the fluence. There is a possibility that it will be done (from clinical data

Chiharu Dermatology Clinic, Dr. Chiharu Watanabe