First time skin microbiome exploration into wrinkle area

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Introduction:

The most external skin layer is alive with many commensal microorganisms which may contribute to maintaining a healthy-looking skin. Skin microbiome is extensively studied in dermatological and cosmetic fields to develop solutions for different skin conditions.
Although the skin microbiome is considered to be an important component of skin health, its relation to the appearance of the skin, and in particular aged and wrinkled skin, is only partially known [1].
We sought to understand the differences of skin microbiota composition between young and aged skin, with a specific focus on wrinkles (crow's feet). We used whole genome sequencing coupled with metabolomic analysis to measure shifts in skin microbiome composition and metabolites.

Materials & Methods:

Informed consent:
Participants signed an Informed Consent Form. Any and all identifying information were treated as protected health information.

Volunteers :
100 Caucasian female volunteers recruited according to their wrinkle grade (WG):
- 50 in young cohort (WG 0-1, 18 to 35 years old)
- 50 in old cohort (WG 5-6, 55 years old or more)

Clinical protocol:

Washout period 2 days (washing the face with water only) → **Swabbing**
- Wrinkle hollow
- Crow's feet & under-eye zone
- Cheek area adjacent to earlobe (control zone)

→ **Microbial DNA study** +  + **Instrumental measurement + Metabolite analysis**

Microbiota analysis protocol:

DNA extraction
- **DNA Extraction** (EpiCentre MasterPure kit)
- **Removal of host DNA** (NEBNext Microbiome DNA Enrichment Kit)

Library preparation
- **Preparation of whole metagenome sequencing libraries** (NEBNext Ultra II FS DNA Library Prep kit)

Sequencing/ Bioinformatic analysis
- **Sequencing** (HiSeq 3000)
- **Human reads cleaning** [2]
- **Taxonomic analysis** (MetaPhlan2)

Metabolites profiling: Combination of GC-MS and LC-MS/MS methods.

Conclusions:

This study gave a deep insight into the **microbiota composition into the wrinkle zone**, confirming some knowledge but also discovering new, such as the unexpected decrease of Lactobacillus namely *L. crispatus* abundance into the wrinkle.
The next steps are to **elucidate the potential function of the species more prevalent in young persons** and how they impact skin.
This understanding may contribute to the development of solutions to favor more desirable and young skin phenotype through either a direct contact with these beneficial bacteria (probiotics) or the use of their beneficial metabolites (postbiotics).

Results & Discussion:

49 subjects in the old cohort and 46 subjects in the younger completed the study.

In the old cohort (*vs* young):

- **A significant increase in alpha diversity** was highlighted by the global analysis including all areas, in agreement with previews studies [3].

- **Significant bacterial shifts were confirmed for:**
  - ***Cutibacterium acnes***, formerly *Propionibacterium acnes,* which had a lower relative abundance on the crow's feet area (37% vs 74%) [3];
  - ***Corynebacterium kroppenstedtii*** which had a higher relative abundance (13% *vs* 2%) [1].

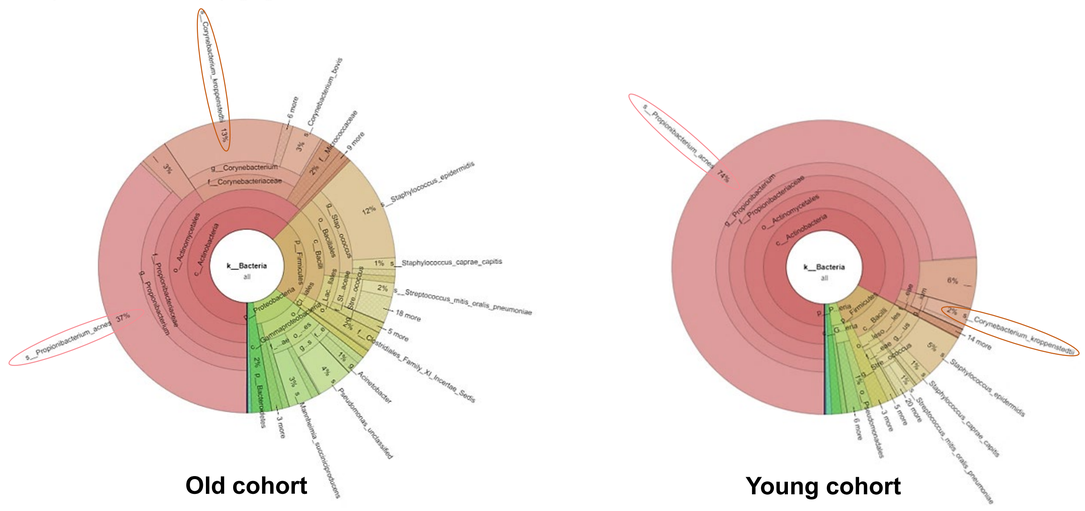

Figure 1: Bacterial composition on the crow's feet area.

- **Lactic acid bacteria abundance and prevalence were decreased in the wrinkle zone and *Lactobacillus crispatus was* the most affected.**

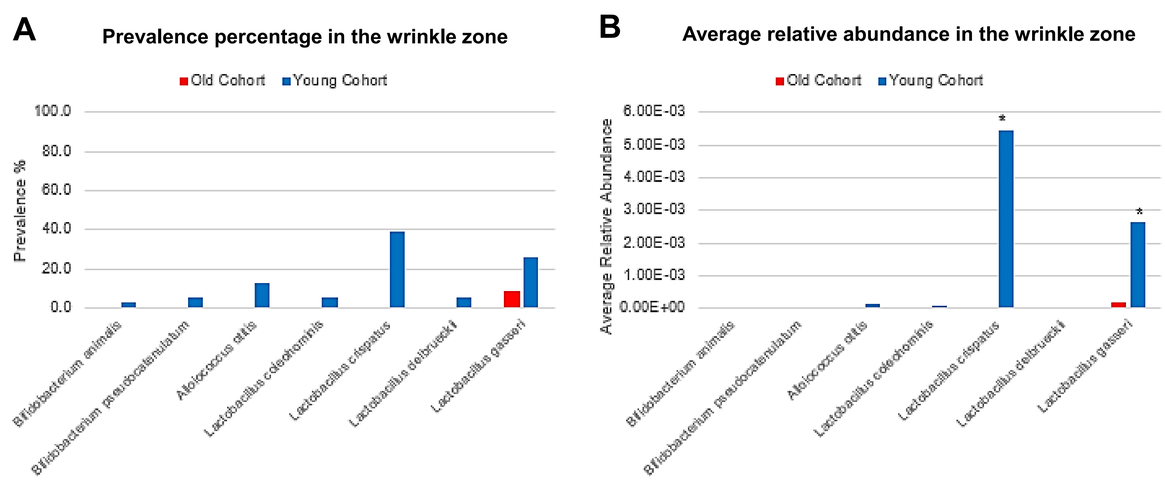

Figure 2: Age-related variation of lactic acid bacteria in the wrinkle zone A- Prevalence percentage and B- Average relative abundance; Statistics: n=33 old group, n=38 young group, Student t test. (*) p<0.1.

- **The metabolomic analysis revealed significant lower levels of fatty acids, lipids and glycerol.**
  - ➔ The decrease of lipophilic strains on aged cohort could be linked to the decrease of lipophilic nutrient on aged skin cohort (sebum, lipids, fatty acid…).
  - ➔ The low amount of glycerol on aged skin could also supports the hypothesis of a lipophilic shift in aging, as glycerol is released from lipase activity.

References:

1-Dimitriu PA, Iker B, Malik K, Leung H, Mohn WW, Hillebrand GG (2019) New Insights into the Intrinsic and Extrinsic Factors That Shape the Human Skin Microbiome. MBio 10:e00839-19
2-Truong DT, Franzosa EA, Tickle TL, Scholz M, Weingart G, Pasolli E, Tett A, Huttenhower C, Segata N (2015) MetaPhlAn2 for enhanced metagenomic taxonomic profiling. Nat Method. 12: 902-903.
3-Shibagaki N, Suga W, Clavaud C, Bastien P, Takayasu L, Lioka E, Kurokawa R, Yamashita N, Hattori Y, Shindo C, Breton L, Hattori M (2017) Aging-related changes in the diversity of women's skin microbiomes associated with oral bacterial. Scientific Reports 7:10567:1-10