MICROBIAL ECOLOGY OF INDIGENOUS FRUITS IN RELATION WITH GEOGRAPHICAL ORIGIN AND/OR PRODUCTION MODE

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Training workshop on Characterisation of Fresh and Processed Fruit Quality
Nong Lam University
Vietnam 23-25th july 2012

Traceability of food commodities

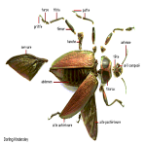
- Traceability of foods (fruits) is only documentary. In case of doubt or fraud, no standardized analysis makes it possible to trace back the origin of the fruit.

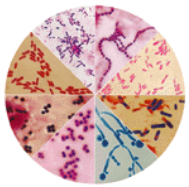
Where do they come from ?

Hypothesis for the determination of origin

The environment has an effect on the micro-flora present on fruit and vegetables

Micro-organisms (bacteria, yeast, moulds)
Insects
Chemical residues
Heavy metals
Isotopes ...

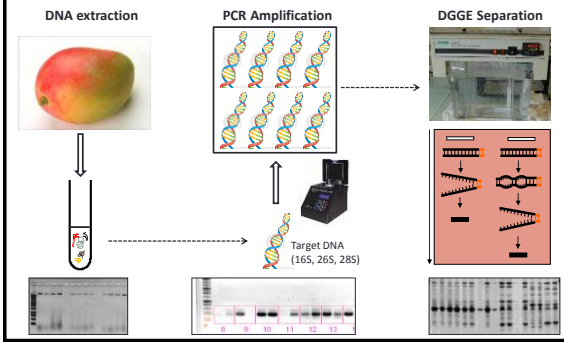




Analyses in Food Microbial Ecology… What for ?

- **Food Safety : Inventory of microbial species associated with food products** (contaminants, pathogens)
- **Food Process/Transformation : Monitoring of the microflora dynamics** (identification of dominant, fermentation flora, etc…)
- **Food quality determinants** (measurable and/or controlled parameters such as pH, Aw, Temperature, Biological activity, toxin levels, Organoleptical compounds, Micronutrients…)
- **Traceability : Microbial Ecology linked to geographical origin and/or production mode of foodstuff**

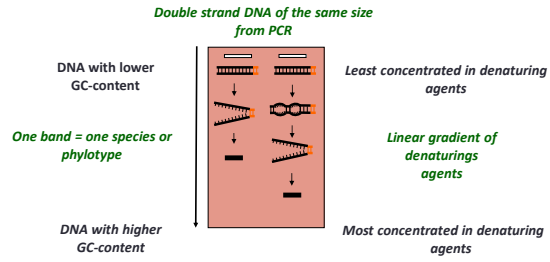
Global Microbial Ecology Analysis using PCR-DGGE technique



Analysis of food microbial ecology at the molecular level (PCR-DGGE)

- Provides a global snapshot of the microbial flora structure
- Culture-independant (no microbe cultivation or isolation)
- Analyses on total DNA directly extracted from foods
- rDNA DGGE profiles : **Food Biological Barcodes** generated by Genetic Fingerprints dependant on the structure of the microbial flora (number et relative abundance of species)

DGGE principle



Applications on various foodstuffs :

- **Fish**
  - **Farm Fish** *from Thaïlande & Vietnam*
    (Ratanaporn Leesing 2005 & Nguyen Doan Duy 2008)
  - **Traditional Fermented Fish** *from Ivory Coast*
    (Clémentine Kouakou 2012)


- **Fruits**
  - **Physalis** from *Colombia, Uganda & Egypt*
  - **Shea tree** from *Mali, Uganda, Senegal & Cameroon*
  - **Clementine** *Spain & Marocco*
    (Aly El Sheikha 2010)
  - **Mangoes** from *Pakistan*
    (Meile 2012)



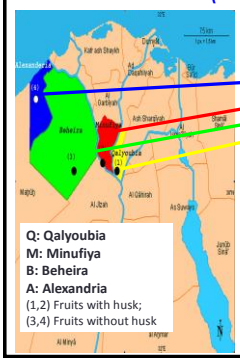
  - **Coffee & Cocoa beans** from *Mexico, Venezuela, Cameroon and Ivory Coast*
    (Noël Durand et Nadège Nganou 2012)

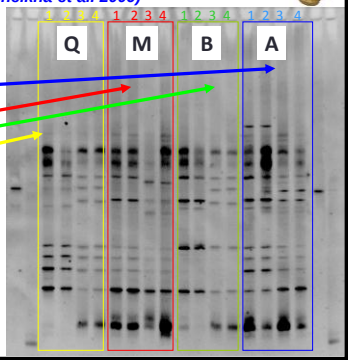

- **Sea salt** from french and portuguese atlantic solar salterns (Meile 2012)

- **Nem Chua** (traditional fermented saussage from Vietnam)

Example : Physalis

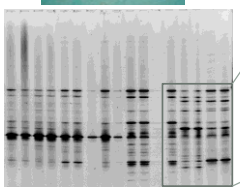
Yeast DGGE profiles of Physalis from 4 regions of Egypt (El Sheikha et al. 2009)

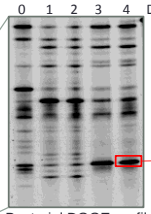




Example of processed food :

Nem Chua (traditional fermented saussage from Vietnam)



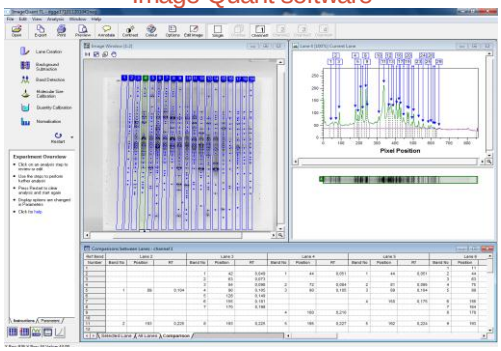


Purification
Sequencing
Identification

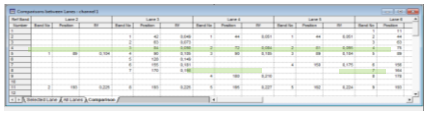
Bacterial DGGE profile
Realized with PHAN THANH TAM
School of Biotechnology and Food Technology, Hanoi University

To be adapted to fermented products such as wine or vinegar

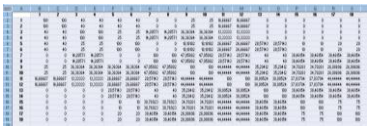
Image Analysis of DGGE profiles using Image Quant software



Comparative analysis of DGGE profiles

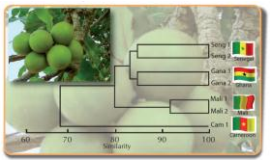


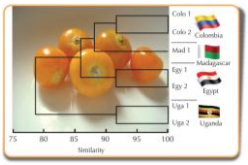
Digitization of the DGGE gel image (Image Quant)

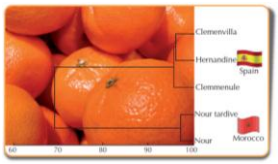


Similarity matrix based on presence/absence of co-migrating bands

Few results obtained with fruits From different origins

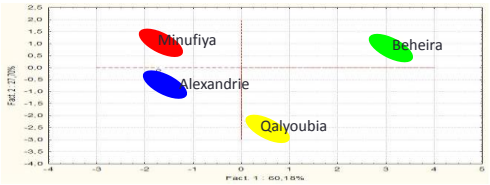


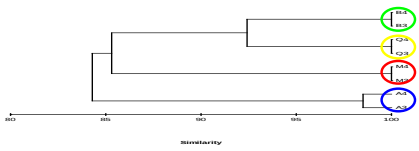




Statistical comparison of **Biological barcodes** (DGGE profiles) allow fruits from different geographical origin to be discriminated

Multifactorial Analysis of data (PCA & Cluster)





Hypothesis on the biological barcode

- Biological barcode influenced by two main factors :

Natural environment (location of production)

Human intervention (agricultural practices)

Comparative analysis of production modes

- Measure the influence of human practices on food microbial ecology
- Example : organic farming as a model
- Discrimination of organic from conventional food products

vs

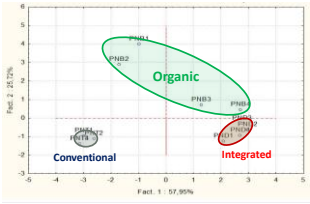
Example with Necarine

- Statistical analysis (PCA)

Nectarines grown using 3 different types of farming :

Conventional, **Organic** and **integrated**

Same variety and location of production



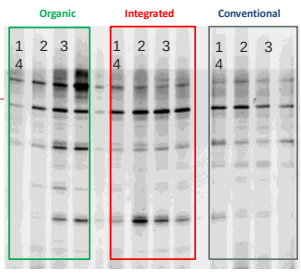
DGGE profiles comparison show that the modes of productions can be discrimnated

EXAMPLE WITH NECARINE

Nectarines produced with 3 different Types of farming were analysed :

Conventional, **Organic** and **integrated**

Same variety and location of production



DGGE profiles using yeast 26S rDNA

Conclusions

- Differences between agricultural practices display a mesurable effect on the global microflora (bacteria, yeast and moulds) of food products
- The geographical origin together with the mode of production provide agricultural products with a unique signature or barcode that can be detected by molecular microbial ecology appraches (such as PCR-DGGE)
- This biological barcode cannot be falsified

Perspectives

- Identify and determine microbial species that could be used as « markers » of geographical origins and/or the production mode
- Strategy for the setup of fast-analysis tools that could be used for authentication and controls along the chain of production and distribution

Aknowledgments

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