

Degree Course: FOOD SCIENCES AND TECHNOLOGIES

Subject: FERMENTATION MICROBIOLOGY

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AIMS

The module of *Fermentation microbiology* aims to give, also by training activities in laboratory, specific knowledge of (a) industrial microbiology and microbial metabolism and of (b) fermented food microbiology, focusing the attention on the selection of microbial starters to use in food field. Moreover, the students will learn some applicative aspects of the microbiology of fermentations; particular emphasis will be given to the microorganism clonal techniques to obtain strains starter for the fermented food production. Throughout the training in the laboratory, adequate emphasis on the strategies to use in order to operate under optimal safety conditions will be given.

PROGRAM

Introductory lesson: presentation of the program contents, the suggested book, and the adopted evaluation modalities. Industrial microbiology: generalities; main process types; microorganisms, substrates and products of the industrial microbiology; classification of metabolites; culture systems and fermentation kinetic; accumulation of metabolites with industrial interest; procedure to select microorganisms with industrial interest: primary and secondary screening; the fermenters; the developing of the fermentative processes; the downstream processing; the main products of the industrial microbiology. Microbial metabolism: the respiration; the fermentation; the regulation of the metabolism. Microbial starter culture: generalities, lactic acid bacteria, *Micrococcaceae*, propionic bacteria, *Bifidobacterium*, acetic acid bacteria, yeasts, moulds; natural/spontaneous fermentations; role of the starters during and at the end of the fermentation; introduction of legislation concerning the microbial starters; preparation of the starter for their inoculation in food; general characteristics of autochthonous microorganisms in food; classification of the fermented food. Salami: generalities, composition, aging, cured pork meat factories; meat microorganisms; natural fermentations and maturation of the salami; drawbacks of the natural fermentation of the salami; preparation and use of the starter cultures for the salami production: *Micrococcaceae*, lactic acid bacteria, moulds and their role in the salami maturation. Dairy products: definition, milk, milk microorganisms, natural fermentation of the milk, kumis, kefir, yogurt; pro-biotic; pre-biotic bacteria; milk fermented with *Bifidobacterium*; milk fermented with *Lactobacillus acidophilus* ("*acidophilus milk*"); pro-biotic yogurt. Cheese: definition, classification criteria, natural fermentation, natural starter; selected starter and use of the starter cultures in dairy industries; fresh, semi-hard, hard, blue-veined, and "pasta filata" cheeses. Wine: definition and composition; microorganisms of the musts; must natural/spontaneous fermentation; drawbacks of the natural fermentation; starter culture; methods of selection; choice of the strain; introduction of the legislation concerning the wine yeasts; preparation and use of the starter in oenology; sparkling wine production; malolactic fermentation. Bread and bakery products: definition, composition, microorganisms of the flour; daily bread; bread with postponed rising; sourdough. Beer: definition, classification, raw material; chemical composition of the barley and malt; preparation of the must from malt; microorganisms of the beer and natural fermentation; starter cultures; malts and acidic musts; *Lambic* beer. Fermented food from leaves and fruits: sauerkraut olives fermented in brine. Extractive fermentation: cacao beans, coffee beans. Oxidative transformations: vinegars. Conclusive lesson: check on the efficacy of the lessons and discussion on related themes; execution of a written test similar to that one that will be given at the exam day; presentation of the laboratory training program.

Laboratory training - Selection of yeast for oenological use. Preparation of the main ingredient (dried grape skins) of the selective medium *grape skin agar*; sterilization of liquid and solid media. Preparation of thermized must - with and without addition of oenological oil and potassium metabisulfite - in tubes and bottles, to evaluate the fermentation vigour and the sulfur dioxide resistance of the yeast strains isolated. Streaking on Petri plates containing Sabouraud agar of tree musts and must-wines samples. Microscopic observation of the following yeasts from food: *Saccharomyces cerevisiae*, *Hanseniaspora guilliermondii*, *Schizosaccharomyces pombe*, *Zigosaccharomyces bailii*, *Candida pulcherrima*, *Candida zeylanoides*, *Debaryomyces hansenii*, *Kluyveromyces marxianus*, *Rhodotorula* sp., in order to acquire the ability to recognize their differences. Microscopic observation of yeast cells from single colonies grown on the plates with *Sabouraud agar*; choice and isolation in slant *Sabouraud agar* of an elliptic strain. Sterilization of liquid and solid media. Inoculation in liquid substrates of standard strains of lactic acid bacteria and acetic acid bacteria stored at -85°C in Protect system. Inoculation of the elliptic yeast strain chosen from the slant *Sabouraud agar* in Petri plates containing *agar acetate*, *agar calcium carbonate*, *BiGGY agar*, and *grape skin agar*; inoculation of the biomass in two test tubes containing thermized must. Evaluation trials of the chosen strain fermentation vigour and resistance to the sulfur dioxide. Inoculation in thermized must. Evaluation of the biomass colour in *BiGGY agar*. Preparation of the kefir; quality judgement of commercial yogurt samples by microscopic observation and evaluation of the ratio cocci/bacilli, length of the chains, presence/absence of eukaryotic cells. Learn the modalities to store microbial cultures by the Protect system; taste of kefir. Calculation of $\Delta 3$ and of the acetic acid production. Microscopic observation of the tetrad produced by an elliptic strains. Calculation of $\Delta 7$. Comparison with $\Delta 3$. Evaluation of the yeast strain behaviour to adsorb/non adsorb colour in *grape skin agar*. **If someone asks, it will be given an English-language summary at the end of each lesson.**

TEXTS ADOPTED

Wessner D.R., Dupont C., Charles T.C. - Microbiologia. Prima edizione. Casa Editrice Ambrosiana (MI), 2015.

Zambonelli C., Tini V., Giudici P., Grazia L. - Microbiologia degli alimenti fermentati. Gruppo Calderini Edagricole (BO), 2001.

Jay J. M., Loessner M., Golden D. A. - Microbiologia degli alimenti. Edizione italiana a cura di Andrea Pulvirenti. Springer-Verlag Italia (MI), 2009.

JUDGMENT

Oral examination in Italian or English language.