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Dr. Ahmed Ouali, 1948-2020

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We were saddened to learn that Dr. Ahmed Ouali passed away on April 4, 2020 after having fought hard against cancer for 10 months. Ahmed was a distinguished meat scientist of the highest calibre and integrity with an outstanding contribution to our understanding of the biochemical mechanisms underpinning muscle to meat conversion by exploring the role of enzymes and their inhibitors. Ahmed was also well known and recognized for his humanist values and for growing them, sharing them and teaching them outside of his professional activities. His fraternal vision of Mankind went far beyond the spirit limits often set by ordinary people; constantly pondering science and consciousness. These values also found a daily place with his family, his students and his co-workers by detecting and often resolving the difficulties which could affect their well-being necessary for them to live and carry out their work in friendliness rather than in falsely imposed competition.

Ahmed Ouali was born on October 4, 1948 in Tizirt, Tizi-Ouzou, Algeria. In 1952, he moved with his parents to Montluçon, France. In 1974, he was trained and graduated with a bachelor's degree in Biochemistry at the University of Lyon. He then, in 1976, earned a joint Ph.D. in Animal Science at the University of Blaise Pascal (Clermont-Ferrand) where he studied at the National Institute of Agricultural Research (INRA, Theix). The title of his doctorate thesis was "The role of muscle proteases on meat tenderization". Subsequently, he was employed in a private laboratory for medical analysis from 1976 to 1978 and thereafter at the Meat Research Laboratory group at INRA, Theix as a permanent researcher. In 1990, he was appointed as a research director and led the "Biochemistry and Functions of Muscle Proteins" unit for 8 years. The Meat Research Station focused their research on many topics including colour and protein oxidation; enzymology and tenderness; and muscle protein functionalities. During his entire

scientific career at INRA, but before his retirement on October 2013, Ahmed was living in Clermont-Ferrand, the city of the famous volcanic chain of the Puy-de-Dôme, with his wife Anne-Marie with whom he had two lovely children: Armelle (41 years) and Gäel (38 years). In 2019, they moved to their new house in Montpellier in the South of France.

During his career as a meat scientist, Ahmed achieved wide recognition for his scientific achievements. He was known for his studies on endogenous muscle proteases and their inhibitors as well as the proteolytic and physiological mechanisms underpinning meat texture determination. He studied most of the proteolytic systems recognised to play a major role in the conversion of muscle into meat, *i.e.* cathepsins, calpains, proteasomes, serine peptidases and more recently caspases. In the early years, Ahmed first started the investigation of muscle lysosomal enzymes and calcium activated neutral proteases on myofibrillar ATPase activity and their role in muscle changes during ageing. He discovered that the sensitivity of the myofibrillar Mg-Ca-enhanced ATPase activity to ionic strength could be an accurate indicator of the degree of meat ageing and called this method the Biochemical Index of Myofibrillar Ageing.

In the middle of 80's and during 90's, he started a series of projects aiming to understand the action of serine/cysteine protease inhibitors considered for years as an enigma by meat scientists. At this summit of his research activities and despite his chronic illness (insulin-dependent diabetes), Ahmed supervised several bachelors, Ph.D. students and postdoctoral researchers, aiming to identify, purify and characterize the endogenous muscle peptidase inhibitors. From a practical point of view, Ahmed's strategy therefore was different from the others as he was looking for the protease inhibitors rather than their target enzymes, *i.e.* serine and cysteine proteases. He first targeted the characterization of low molecular weight bovine muscle cysteine protease inhibitors. Further fractionation of muscle crude extracts using many novel chromatographic techniques designed by Ahmed, (for which he was a well-known actor at this time), allowed him to reveal three major trypsin inhibitory fractions differing in their molecular mass (F1-Mr: 50–70 kDa; F2-Mr: 40–60 kDa and F3-Mr: 10–15kD) which were later analysed for years to come. These allowed him in the 2000's after much work on bovine muscle 20S proteasome, to discover for the first time that the F1 and F2 fractions comprised of a large set of closely related trypsin inhibitors encoded by at least 8 genes *bov-SERPINA3-1* to *A3-8*. These fractions were found to inhibit strongly in a cross-inhibition manner initiator and effector caspases, a group of cysteine proteases orchestrating the demolition phase of apoptosis. Further work identified that these inhibitors all belong to the SERPIN superfamily, known to form covalent complexes with their target serine and cysteine proteases and located within muscle cells and found in all tissues and fluids examined irrespective of the animal species. Taken together, these results allowed Ahmed to build a new hypothesis in the field of meat science by ascertaining the involvement of new pathways including the role of heat shock proteins in the *post-mortem* conversion of muscle into meat.

Indeed, one of Ahmed Ouali's major contributions for which he will be remembered in his field of research is the theory of the onset of programmed cell death or apoptosis in *post-mortem* muscle foods and the pivotal role that it plays during tenderization. His elegant prospective in 2006 after many years of devoted research in his laboratory and knowledge acquired since 1995 on apoptosis especially in humans, allowed him to emphasize that apoptosis can constitute an exciting and revolutionary hypothesis about the biochemical mechanisms involved in the conversion of muscle to meat. This was published in *Meat Science* journal in 2006 following an invited keynote at 52th ICoMST held by Teagasc in Dublin. This hypothesis allowed lead to a new exciting era of research, supported by the use of OMIC technologies, on the pathways behind the conversion of muscle into meat by integrating an additional phase before the rigor process corresponding to the apoptotic phase. At the same time, Ahmed directed other projects which aimed to identify biomarkers of meat quality. He was convinced that the quest for biological predictors of tenderness and other qualities of the meat is an absolute necessity, in

order to enhance valorisation of carcasses by directing them shortly after slaughter towards an optimal use on the basis of their potential qualities. He further collaborated with colleagues from other universities, namely the Cell Motility Laboratory from the “École Pratique des Hautes Etudes”, University of Montpellier, to study the calpains localisation and their interaction with titin and received several years funding to conduct his research from the French Muscular Dystrophy Association. Ahmed also kept strong collaborative links with Algeria, namely with the Universities of Constantine and Algiers.

Ahmed was well known for his welcoming and inclusive spirit, deep sense of fairness, generosity, and complete lack of self-importance. He trained many bachelors, Ph.D. students and postdoctoral researchers, mentored a number of young scientists coming from all over the world (Algeria, Spain, Morocco, Tunisia, Mexico, Italy, Germany, Ireland ...) and was also eager to transfer his knowledge to his colleagues in different INRA research units and around the world. Along with his rectitude and honesty, the most distinctive feature of his personality was his unbound passion for muscle and meat biochemistry, a science to which he dedicated a great part of his life with priceless enthusiasm and generosity. Indeed, Ahmed wrote numerous books, book chapters, patents and comprehensive reviews in his field of expertise in addition to highly cited research articles. Ahmed participated in many national and international meetings as an invited keynote international speaker such as ICoMST.

We shall, from now on, remember and associate Ahmed with major scientific contributions to Meat Science, but he was mostly known as a modern day dervish for his humility, wisdom and with no concern for financial rewards or notoriety in a very competitive world.

We hope that Dr. Ahmed Ouali would find this letter a fitting tribute to the meat scientist that we should all aspire to be. From now on, the best we can do is to continue his work in Meat Science following his principles of not compromising on excellent science and demonstrating the best of humanity.

To Ahmed, our gratitude and eternal remembrance! You are dearly missed!

Conflicts of interest

There are no conflicts of interest.