Rainbow trout white muscle proteome: changes due to slaughter stress

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Proteomics has been largely used to study muscle tissue. This tool allows the identification of many muscle specific proteins and was shown to be efficient in comparing various physiological and *post mortem* conditions in relation with meat quality. In fish, several studies also demonstrated the interest of proteomics tools to identify muscle protein changes in various conditions.
Fish flesh texture is one of the main organoleptic qualities for product acceptability. Fish flesh texture is related to muscle three-dimensional organization (structure) but is also determined by muscle components among which proteins are the most important.
Amongst factors demonstrated to affect fish texture, the conditions of animal slaughter are determinant. Stress at slaughter, associated to an intense muscle exercise was shown to drastically affect flesh texture. This is associated with changes in *post mortem* metabolism, for example a faster fall of muscle pH due to muscle glycogen depletion leading to a faster entry in *rigor mortis*. Intense pre-slaughter muscle exercise was also shown to enhance *post mortem* proteolysis, all of this contributing to a softer flesh texture. In this context, a proteomic approach was chosen to identify new markers that can explain changes in fish texture associated to slaughter conditions.
Two groups of fish were constituted. The control group was slaughtered in limited stress conditions (rapid fishing + anesthesia), and the other group was slaughtered after 15 min of confinement stress associated with an intense muscle exercise. Deep white muscle from 5 fish of both groups was sampled within 45 min after death, frozen in liquid nitrogen and stored at –70°C. Extracted proteins (Urea 8M, Chaps 4%) were separated by IEF on IPG dry strips 3-10NL followed by SDS-PAGE on 12.5% acrylamide gels. Three gels for each sample were made. 500 spots were analysed for differences due to the slaughter method. Different strategies for the analysis of image gels were tested to take into account variability due to the method and to the animal. Depending on the method of analysis, 20 to 40 spots were found to be differentially expressed according to the slaughter conditions. A total of 215 different spots were excised and submitted to in-gel trypsin digestion. To date, 110 samples were submitted to MALDI-tof mass spectrometry and ≈ 50 of them were successfully identified. Most of the identified proteins were specific muscle structure proteins (actin, myosin light chains, tropomyosin, troponins) and metabolic enzymes. Only two of the spots differing between slaughter conditions were to date identified as capZ and alpha-enolase.

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