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Analysis of skeletal muscle tissue expression profiles in pig to identify genes involved in meat quality traits: effect of stress conditions before slaughtering in different pig breeds

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ABSTRACT

The knowledge of genes and molecular processes involved in the stress susceptibility and reaction could be important for the improvement of meat quality traits. The aim of the research was the identification by microarray analysis of skeletal muscle genes differentially expressed in pigs divergent for meat quality. The expression profiles were studied in subjects exposed or not exposed to a physical stress (a run in controlled conditions) before slaughtering. *Semimembranosus* muscle samples were collected at slaughtering and stored at -80°C before RNA extraction. These samples were obtained from 38 unrelated castrated male pigs belonging to 3 different breeds and reared up to 120 kg in the University of Bologna experimental farm: 10 Italian Duroc and 10 Italian Large White pigs with genotype CC at the *RYR 1* 1843 polymorphic site and 18 Pietrain pigs (CC, CT and TT in the not stressed group and only CC and CT in the stressed group). Microarray analysis was performed using two platforms: a cDNA based microarray produce by the University of Aarhus (~27,000 porcine cDNAs) and the Operon/Qiagen pig 11k Oligo set (version 1.0) (10,665 70mers oligos). Total RNA extracted from the samples was reverse transcribed, labelled with fluorescent Alexa Fluor 555 dye and individually hybridized to both cDNA and Oligo microarrays. The labelled samples were hybridised together with a common reference sample consisting of Alexa Fluor 647 labelled total RNA extracted from skeletal muscle and heart tissue from a pig not related with those of the experiment. ScanArray Gx (Perkin Elmer) scanner and ScanArray Express software were used for image acquisition and data normalization. ANOVA has been performed without to include 3 Pietrain pigs not stressed with genotype CC at *RYR 1*. Using the Mixed procedure of SAS that consider fixed and random factors the adjustment for overall effects of array and dye across genes was performed. The residuals from this normalization process were used to test the effects of the factors included in the model on gene expression. Differences in the least square means were obtained and tested for significant differences by using a t-test. The false discovery rate was used to account for multiple testing. A false discovery rate of 5% was accepted. Preliminary results of the comparison between stressed and not stressed pigs obtained with the Oligo array showed that more than 20% of the genes upregulated in the group of stressed pigs belong to functional categories involved in regulation of cellular physiological processes. The first results from the comparison between treatments within breed seems to evidence differences among breeds in stress response and can provide indication on potentially interesting genes influencing pig meat quality.