

## SUMMARY

### *Use of Amiata breed donkey milk in allergic and / or intolerant children: nutritional and sanitary characterization for food safety*

Key words: donkey milk, hygienic production, physico-chemical composition, microbiological quality

The aim of this study was to provide elements of characterization of the sanitary and nutritional criteria for the use of donkey milk (DM) as a product for human consumption, these criteria concern the entire production chain from plant-structural, animal health up to the hygiene requirements of milk.

A series of preliminary evaluations carried out by the various Operating Units (U.O.) have allowed to better understand the DM product, from the point of view of production (breeding, animals, milking ...), tolerability and palatability at consumption, nutritional characteristics, sanitary parameters at production, conservation and transport phases and up to regulations.

In Tuscany there is the autochthonous Amiata breed and a recent project, realized with 2007-2013 funds from the Tuscany Region's PSR, has allowed the creation of an innovative production chain for the production of DM, located and implemented at the Regional Forestal Agricultural Complex "Bandite di Scarlino", a partner of this project that has been recognised according to EU Reg. 853/2004. At this farm there are about 160 donkeys, bred in very large paddocks in semi-wild state ad libitum hay fed and with a commercial concentrated pellet administration limited to the milking phase. The group consisting of lactating donkeys (30), was homogeneous by lactation age and apparent health status and was subjected to monthly milking and clinical controls for a period of 16 months. Blood and faecal samples and diagnostic swabs of the genital tract were also performed in order to determine animal health parameters, welfare and general assessment (body condition score). The automatic milkings were made twice daily three hours after separating the foals and the main measurements carried out on top of quantity of milk produced were fat, proteins, lactose, caseins, freezing point, lean dry residue, pH and urea. Total bacterial load and somatic cell counts were the main value considered for the evaluation of the production process; the measurements were conducted using Bactoscan FC and Fossomatic 5000.

Additional milk individual and mass samples were also taken for qualitative assessments of milk and for research of mastitis causative bacteria. The analyzes were conducted according to microbiological laboratory procedures and in agreement with the "Laboratory handbook on mastitis council" (National Mastitis Council, 1999). Other analyzes on milk samples have evaluated the presence of mycotoxins according to the analytical methods used in the laboratories.

The parasitological analyzes from faecal samples were carried out within 24h from the collection (rectal ampoule and sterile containers) according to current laboratory methods for the qualitative / quantitative research of enteroparasites and respiratory parasites.

Cervical swabs were performed before the start of the trial aiming to investigate the (asymptomatic) presence of the main pathogens of the equine reproductive system. The exams were carried out according to the accredited microbiological diagnostic laboratory procedures and in accordance with the OIE manual.

Blood samples from the jugular vein were made with vacutainer, stored at + 4 ° C and subsequently separated by centrifugation and frozen at -20° C. In addition to the clinical evaluation by inspections, measurements were made of haemochromocytometry with formula, metabolic parameters of hepatic and renal function and of energy metabolism in relation to diet and production needs.

The milk has also been analyzed from the nutritional and nutraceutical point of view, mainly for the content of the lactoprotein and lipid.

Clinically the animals have shown values in the ranges of physiological parameters of donkeys.

There was no serological positivity to the main pathogens of donkeys or zoonotic, both viral and bacterial.

A moderate prevalence of *streptococcus equi zooepidemicus* and *klebsiella pneumoniae* (9% and 6%, respectively) was evident from the cervical swabs. These pathogens were not shown clinically: it seems to represent the data of their commensality for the urinary tract that only under certain stress conditions can worsen with clinically evident forms.

Intestinal and respiratory parasites were found with a prevalence of 96% and 19% respectively, no hepatic parasites were detected. The average number of enteroparasites was 886 eggs per g. of feces (*Strongiloides*), highlighting the need to undertake a control plan of such parasites, limiting as much as possible the use of pharmaceutical molecules in production animals. It is important to note that both for intestinal and respiratory parasites there are no significant clinical manifestations.

Mastitis infections were also moderate, confirming the marked natural antibacterial activity of donkey milk: *staphylococcus aureus* and *streptococcus equi zooepidemicus* with a prevalence of 3 and 6%, respectively have been isolated from two individuals.

Dry matter value corresponds to bibliography data. The protein and the lactose content of donkey milk are closer to the values of human milk than to cattle. The lower content of proteins and caseins compared to cow's milk make the DM more suitable for consumption for the categories of allergic people and with problems of milk digestibility. The lactose is more present in DM than in cow's milk, it confers a good palatability of the product and stimulates the absorption of calcium in the intestine, this determines the geriatric use of DM.

The lysozyme activity characteristic of donkey milk is very similar to that of human milk and much higher than bovine milk (only a minimal presence); we recall that lysozyme and lactoferrin are the most responsible for the antibacterial activity of milk. The low amounts of fat and the morphometry of the fat molecules make donkey milk important also for diets needed in obesity or for people with cardiovascular problems. The smaller sizes of donkey milk fat globules compared to those of cow's milk, make the first more digestible and with a greater contribution of bioactive elements and polyunsaturated fatty acids. The ratio of unsaturated / saturated fatty acids in donkey milk is more similar to human than in cattle.

The animals have shown good health conditions and no evidence of pathological conditions. The load of parasites must be taken into consideration in order to avoid that an overload can reduce the production. The production processes have not highlighted any particular criticality from the hygienic point of view even if the sporadic and preliminary isolation of *Staphylococcus aureus* makes it essential to carry out checks to ensure compliance with the provisions of current legislation. DM is characterized by a high lactose content, low fat and protein content and high content of unsaturated fatty acids. The unsaturated fatty acids and, in particular, omega3 give the product a particular health benefit for targeted diets for humans in situations of intolerance, allergy to cow's milk, in situations of dyslipidemia, in prevention of cardiovascular diseases, in hypolipidemic diets and in calcium-free geriatric diets.