success of ART techniques. EcoFoodFertility Project

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Introduction: Protein p53 is well known as "The guardian of genome"; it changes its concentration in human spermatozoa DNA in relation to the damage of the latter. It has been suggested that the role of the p53 ancestral gene was to ensure the integrity of the genomic germline and the fidelity of the development process. The aim of this study is to evaluate if different concentrations of p53 protein in human spermatozoa could influence embryo quality and pregnancy rate and possibly representing a potential predictive marker of sperm quality for successful fertilization.

Methods: From July 2013 to June 2017 we have examinated retrospectively 79 couples with 2-5 years of infertility history. Males had an average age of 27 ± 7,5 years, sperm concentration of 33,8 ± 6,2 mil/ml, progressive motility of 41,4 ± 8,3 and a typical morphology of 16,5 ± 3,5 according to Kruger's method. We have divided the couples on the basis of p53 levels: Group A: 0,35–1,65 ng/mil (21 males); Group B: 1,66–3,57 ng/mil (32 males); Group C: 3,58–14,53 ng/mil (26 males). We have evaluated the number of embryos at stage of 6–8 cells, obtained at the third day of embryo development, in these three different group. In order to evaluate the concentration of p53 protein, we first proceeded to a DNA extraction with forensic method and then to a quantification p53 protein with ELISA-immunoenzymatic assay, expressed in ng/million of spermatozoa.

Results: We have observed different percentage of embryo development at stage of 6-8 cells in the third day and different pregnancy rate (PR):Group A: 101 embryos at 6-8 cells/ 147 total number of obtained embryos in this group (68,4%) and PR = 52,38%. Group B: 128/240 (53,5%); PR = 37,50%; Group C: 79/216 (36,1%); PR = 7,69%. These results support the hypothesis that an high concentration of p53 in human sperm DNA is associated to a low percentage of embryos able to reach the stage of 6-8 cells in the third day of development and also to a lower pregnancy rate. So p53 levels can be considered as a predictive value to embryo development and pregnancy rate.

Conclusions: Protein p53 is a sequence-specific transcription factor that responds to a wide variety of stress signals (environmental insults and bad lifestyle) as we are investigating within the ecofoodfertility project. Particularly quantitative research of p53 could be considered as a novel biomarker of sperm quality, able to predict the success of ART techniques, and could open a new road for infertility diagnosis.

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p53 protein in human spermatozoa. (Preliminary data. EcoFoodFertility Project)

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Introduction: Oxidative stress has been identified as one of the many mediators of male infertility by causing sperm dysfunction. The aim of this study within the preliminary diet strategies of Eco-FoodFertility project is to evaluate the variations of p53 protein after the introduction of an antioxidant in the daily diet and changing the life-style.

Methods: We recruited 45 male participants (age 28.0 ± 5.6) with sperm concentration (4 to 50 Million/Millilitre), Motility (2 to 3+), morphology upper 14%. The spermatozoa DNA damage were evaluated on the quantitative determination of p53 protein. We realized a DNA extraction with forensic method and a successive quantification with ELISA-immunoenzimatic. The values were expressed in ng/Million of spermatozoa. The participants were divided in 3 groups according to the different p53 protein concentration at time 0′: Group A, 20 patients with [p53] included between 3,68 and 5,7;Group B, 14 patients with [p53] included between 6,0 and 10,96;Group C, 11 patients with [p53] included between 11,02 and 17,85. We proposed to the participants to integrate to the principal meals (breakfast, lunch and dinner) 400 mg of alpha – tocoferol for 30 days. After 30 days we subjected the participants to seminal fluid analysis to evaluate the p53 protein concentration. After 3 months without taking medicine that could influence the state of semen, we proposed to members of the group B and C to take part to a new observational study adding the alpha-Tocoferol in the diet for 30 days with indications about life style regarding have meals at regular time, eliminating some food and drinks that have a strictly contact with plastic and a moderate physical activity 2–3 times a week.

Results: After 30 days: Group A: values at time 0' of 5,69 ± 2,01, values after 30 days 1,25 ± 0,4 with a reduction equal as 71,01%. Group B: values at time 0' of 8,48 ± 2,48, values after 30 days 4,71 ± 2,16 with a reduction equal as 42,11%. Group C: values at time 0' 14,43 ± 3,41, values after 30 days 12,11 ± 2,87 with a reduction equal as 14,16%. Using the method of the first study, after 3 months: Group B: new determination of [p53] at time 0' is 8,57 ± 2,45, values after 30 days 3,29 ± 2,02 with a reduction equal as 59,5%. Group C: new determination of [p53] at time 0' is 14,15 ± 2,84, after 30 days 11,01 ± 2,77 with a reduction equal as 23,30%.

Conclusions: Introducing alpha-Tocoferol in the diet, we have a substantial decrease of p53 in the group "A". Adding advices to the life-style of groups "B" and "C", reduction of p53 is more marked and, in some cases, values are normalized. The synergic