COMMITTEE PAPER FOR DISCUSSION **ACNFP/74/8**

ADVISORY COMMITTEE ON NOVEL FOODS AND PROCESSES

EFFECT OF GM SOYA ON NEWBORN RATS

Issue

The Committee is asked to consider the results from a preliminary study on the effect of herbicide resistant soya on pregnant and new-born rats and to advise on what conclusions may be drawn from this work.

Background

1. In mid-October 2005, results from a rat feeding study were published on the Internet by a Russian organisation, the National Association for Genetic Security. The report (Annex 1) explained that the study had been presented to the Association's symposium on genetic modification on 10 October by Dr Irina Ermakova, a scientist at the Institute of Higher Nervous Activity and Neurophysiology at the Russian Academy of Sciences.

2. The study involved feeding groups of 3 rats on a standard diet supplemented with soya flour from conventional and genetically modified sources. Further details of the study design and the results were obtained from the author (Annex 2). The GM soya is "Roundup Ready", which is the trade name for glyphosate tolerant soya developed by Monsanto. The identity of the GM and non-GM soya was confirmed by genetic analysis.

3. The soya flour was not incorporated directly into the diet but was offered to rats on a small plate, mixed with water. The rats were initially housed groups of 3 and given 20 grams of soya flour each day. Around the time of delivery, the rats were housed individually and given 5 to 7 grams daily, with the addition of 1 gram of soya for each pup (I.e. 16-18 grams for a mother with 11 pups). Results of chemical analysis of the soya flour may be available in time for the Committee meeting.

4. Food ingredients derived from glyphosate-resistant soya containing the event 40-3-2 were evaluated by the ACNFP in 1994 when the Committee concluded that the products were as safe for human consumption as other conventional soya bean products. UK clearance was given in February 1995 (prior to the introduction of EU-wide regulations on GM foods.

Results and discussion

5. The results indicate that the litter sizes were similar in the rats given GM soya, compared with those given non-GM soya and a control group given only commercial feed pellets. However, there were significant reductions in the survival and body weights of the offspring in the GM soya group. This is a small-

scale study and these findings are not accompanied by results of investigations into the causes of death or other clinical parameters.

6. Reproductive studies are not required as part of the standard evaluation of GM and other novel foods. However, a study in mice with GM herbicide-resistant soya was published in 2004 and found no adverse effects (Annex 3).

Committee Action Sought

7. The Committee is asked to consider the results from this preliminary study and to advise on conclusions that may be drawn regarding the safety of GM herbicide-resistance soya.

Secretariat

November 2005

Annexes attached

Annex 1: Internet report: "Genetically modified soy affects posterity: Results of Russian scientists' studies".

Annex 2: Influence of genetically modified organisms on posterity of rats: preliminary studies (paper by Dr Irina Ermakova).

Annex 3: "A generational study of glyphosate-tolerant soybeans on mouse fetal, postnatal, pubertal and adult testicular development" Brake DG and Evenson DP; Food and Chemical Toxicology 42 (2004) 29-36.

ADVISORY COMMITTEE ON NOVEL FOODS AND PROCESSES

Internet report: "Genetically modified soy affects posterity: Results of Russian scientists' studies".

Secretariat
November 2005

Article from website of the (Russian) National Association for Genetic Security

http://www.geneticsafety.org/info.php?txt_id=19&nid=776 (English version of their original article)

Genetically modified soy affects posterity: Results of Russian scientists' studies

On October 10, during the symposium over genetic modification, organized by the National Association for Genetic Security (NAGS), Doctor of Biology Irina Ermakova made public the results of the research led by her at the Institute of Higher Nervous Activity and Neurophysiology of the Russian Academy of Sciences (RAS). This is the first research that determined clear dependence between eating genetically modified soy and the posterity of living creatures.

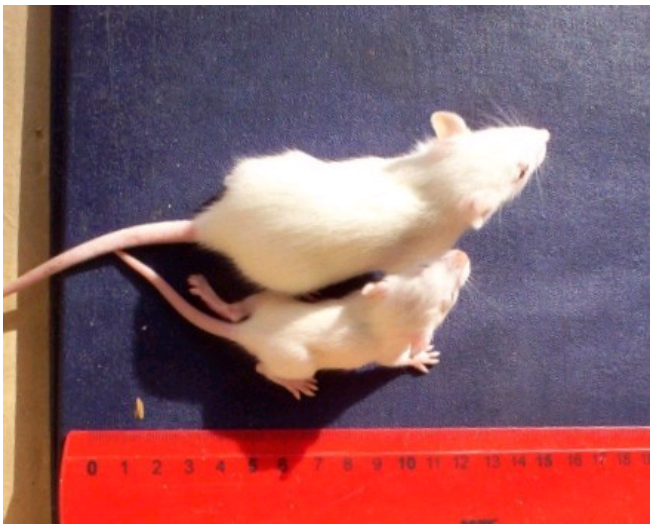
During the experiment, doctor Ermakova added GM soy flour to the food of female rats two weeks before conception, during conception and nursing. In the control group were the rat females that were not added anything to their food. The experiment was formed by 3 groups of 3 female rats in each: the first one was control group, the second one was the group with GM-soy addition, and the third one with traditional soy addition. The scientists counted the number of female species to give birth and the number of born and died rats.

After the result of the first stage, the second part took place. Now the rats were divided into two groups – one with GM-soy addition in their food, and other without the GM-soy. In three weeks the scientists received following results:

Additions	Female that gave birth	Born rats	Dead rats (in three weeks)	Percent of dead rats	Rats still alive
Control group	4 (of 6)	44	3	6,8%	41
With GM-soy	4 (of 6)	45	25	55,6%	20
With normal soy	3 (of 3)	33	3	9%	30

Thus, according to these results, the abnormally high level of posterity death has been detected at the posterity of the female species with GM-soy added to their food. And 36% percent of born rats weighed less than 20 grams that is an evidence of their extremely weak condition.

«The morphology and biochemical structures of rats are very similar to those of humans, and this makes the results we obtained very disturbing,» said Irina Ermakova to NAGS press office. According to NAGS Vice-president Aleksey Kulikov, the data received by Dr.Ermakova confirm the necessity of full scale tests of GM-products influence over living creatures.



On the photo: the results of the experiment. Same age rats from the control group (on the left) and the «GM-soy» group.

ADVISORY COMMITTEE ON NOVEL FOODS AND PROCESSES

Influence of genetically modified organisms on posterity of rats: preliminary studies (paper by Dr Irina Ermakova).

Secretariat
November 2005

INFLUENCE OF GENETICALLY MODIFIED ORGANISMS ON POSTERITY OF RATS: PRELIMINARY STUDIES.

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I. INTRODUCTION

The hazard of genetically modified (or transgenic) organisms was shown for Human health and Environment. The term "Genetically Modified Organisms" (GMO) refers to plants and animals with genes transferred from other species in order to produce certain characteristics, for example, such as resistance to pests and herbicides.

Transgenization (via pollen, seeds and organic rests) leads to the genetical pollution of Environment, to the decrease of biovariety. The scientists repeatedly warned about dangerous of GMO for human and the Environment (Traavik, 1995; Ho and Tappeser B., 1997; Pusztai, 2001; Chirkov, 2002; Kuznetcov et al., 2004; World Scientists Statement, 2000 and many others). Four main sources of hazards and problems of GMO declared in World Scientists' Statement (Supplementary Information on the Hazards of Genetic Engineering Biotechnology): 1) those due to the new genes and gene products introduced; 2) unintended effects inherent to the technology; 3) interactions between foreign genes and host genes; 4) those arising from the spread of the introduced genes by ordinary cross-pollination as well as by horizontal gene transfer.

"Artificial" genes issued in the Environment, like a virus, can insert into genome of cells of all species of plants and animals, including the man. This process called "horizontal moving" of genes (moving from species to species) already has resulted in occurrence of new viruses and bacteria, causing sharp toxicosis, autoimmune reactions, tumor diseases.

Consumption of GM-food by animals leads to the negative changes in their organisms. Experiments, conducted by A. Pusztai (1998), showed that potatoes modified by the insertion of snowdrop and jackbean genes that code for insecticidal proteins stunted the growth of rats and damaged their immune system. Feeding rats with baked transgenic potatoes significantly affected some of their vital organs including the kidneys, thymus, gastrocnemius muscle and others (Pusztai, 1998). These data were confirmed by independent panel of 23 scientists (Peer review, 1999). Similar effect of GM-potatoes on rats was obtained at Institute of Nutrition in Russia (from Centre of public information, 16 December 2004; Ermakova, 2005a). In his article "Genetically Modified Foods: Are They a Risk to Human/Animal Health" (2001) A. Pusztai asked**:** "How can the public make informed decisions about GM foods when there is so little information about its safety?" He considered that the main reason of the informative scarcity was the difficulties to evaluate the safety of crop-derived foods comparing only with the individual chemical, drug, or food additives.

It is important to know two standart methods usually used to introduce a new DNA (gene) into the cell of a plant which is going to be modified: the "shot-gun" technique and Agrobacter tumefacien method. Both methods are not perfect and don't guarantee the safety of the GM-organisms created with their help for the Environment (Kuznetcov et al., 2004; Wilson et al., 2004; Ermakova, 2005b).

The supporters of GMO assert that the GM-components are completely destroyed in the digestive tract of men and animals. However rings of plasmids are steadier than it was believed against the digestion. Plasmid DNA and GM-DNA were found in microflora of intestine and in saliva in man (Mercer et al., 1998; Coghlan, 2002). Experimental researches in mice showed that ingested foreign DNA can persist in fragmented form in the gastrointestinal tract, penetrate the intestinal wall, and reach the nuclei of leukocytes, spleen and liver cells (Schubbert et al., 1994). Important data were obtained when a plasmid containing the gene for the green fluorescent protein (pEGFP-C1) or bacteriofaphage M13 DNA was fed to pregnant mice. Foreign DNA, orally ingested by pregnant mice, was discovered in blood (leukocytes), spleen, liver, heart, brain, testes and other organs of foetuses and newborn animals. The foreign DNA was detected in association with both chromatids (Schubbert et al., 1998). They considered, that maternally ingested foreign DNA could be potential mutagens for the developing fetus.

GMO with modified genetic material are not found in nature under natural conditions. It is necessary to know the interactions of GMO with alive organisms and influence on them. However there is a lack of investigations of GMO influence on plants and animals and, first of all, on mammals and their posterity.

Investigations of the influence of GM-soya (Roundup Ready, RR) on posterity of Wistar rats were performed in our experiments.

II. METHOD

Females Wistar rats weighting 180-200g received additionally to the diet the soya flour (5-7 g for each rat) in their home cages two weeks before mating, during mating, during pregnancy and more soya during lactation: traditional soya (Trad. soya) and GM-soya (Roundup Ready, RR). The scheme of experiments was the following: females received dry pellets as diet in the special place on the top of cage. Rats of two groups additionally received soya flour mixed with water in the special small plate put inside the cage. Three females of each group were kept together in the cage before delivery. Three rats from soya groups received about 20g soya + 40 ml water, so about 5-7g flour for each rat. Before delivery the pregnant rat was placed into the individual cage and was given 5-7g plus 1g soya for every kid after birth. When pups grew up and could eat soya themselves, the doze of soya was increased till 2-3g for every pup. All rats had free approach to the soya. The scheme for trad soya was strictly the same as for GM-soya. Usually all rats ate the soya very well.

Group of rats, which received only diet without any food additives, was considered as the Control group. The weight and mortality of rat pups were analyzed.

III. RESULTS

The high mortality (~ 55,6%) of rat pups in first generation after addition of GM-soy (Roundup Ready, RR) into the diet of rat females (before mating, during mating, pregnancy and lactation) in comparison with control group (6,8%) and Trad. soya group (9%) was revealed in these experiments (Table 1). The high mortality of pups was noticed in all females from GM-soya group (Tabl.2). Most of rats died during first week (tabl.3). Weight of 36% rat pups, whose mothers were fed by GM-soya, was less than 20 grammas in two weeks after birth in comparison with control group and group Trad. soya (6% and 6,7% accordingly) (Tabl.4). Females and survived rat pups, which began to eat the GM-soya themselves, didn't die. Females and pups, fed by GM-soya, were very aggressive: they attacked and bit each other and the worker, who took care about them.

Table 1. MORTALITY OF RAT PUPS (data in three weeks after birth)

Groups	*Delivered females*	*Number of newborn rats*	*Number of died rat pups*	*Died rat pups in %*
Control	4 (from 6)	44	3	6.8%
Trad. soya	3 (from 3)	33	3	9%
GM soya	4 (from 6)	45	**25**	**55.6%**

Table 2. NUMBER OF DIED RAT PUPS FROM GM-SOYA GROUP.

Females	*Number of newborn rats*	*Number of died rat pups*	*Died pups in %*
1 female	11	7	64%
2 female	8	4	50%
3 female	13	6	46%
4 female	13	8	62%

Table 3. Distribution of rat pups weight in two weeks after birth.

Groups	*50-40 g*	*40-30 g*	*30-20 g*	*20-10 g*
Control	12.5%	37.5%	44%	6%
Trad. soya	0%	20%	73.3%	6.7%
GM soya	0%	23%	41%	**36%**

Table 4. Time of death of rat pups from all groups.

Groups	*1st week*	*2nd week*	*3rd week*
Control	4.5%	2.3%	0
Trad. Soya	9%	0	0
GM-soya	**31,1%**	**13,4%**	**11,1%**

DISCUSSION:

We supposed that the negative effect of GM-soya on newborn pups could be connected with the IMPERFECT agrobacter Tumefacien's method by using plasmids with foreign genes, which could lead to the penetration of foreign genes or "unstable gene construction" into sexual cells or\and into cells of fetus according to investigation of Schubbert et al.,1998. The instability of gene construction was described for GM-soya by Windels et al., 2001, and for rice by Yang et al., 2005. Negative effect of GM-soya could be mediated also by the accumulation of Roundup in GM-soya. In order to understand the mechanism of GMO influence on mammals and their posterity it is necessary to perform complex researches, including histological, biochemical, genetic, embryotoxicological and physiological investigations.

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