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Letter to the editor

Comment on "Long term toxicity of a Roundup herbicide and a Roundup-tolerant genetically modified maize" by Séralini et al.

On 19 September 2012, Séralini et al. published online in this journal the results of a long term toxicity study that, in their opinion, revealed clear indications that genetically modified NK603 maize and Roundup are dangerous to health. VIB has reviewed the Séralini et al. paper in detail (VIB, 2012) and in this comment, we point out the most critical flaws in Séralini's design, interpretation and reporting. Because of these fundamental flaws, the conclusions of Séralini et al. are not substantiated in any way.

Seralini et al. fed rats during a period of two years with either NK603 maize (three different doses: 11%; 22% or 33%), with NK603 maize (the same three different doses) that was sprayed with Roundup, or provided drinking water in which Roundup was present (three different concentrations: 50 ng/l, 400 mg/l or 2.25 g/l). In this way nine different treatments were tested and were compared with only one negative control, being a diet with one dose (i.e. 33%) of near-isogenic non-GM maize. Each diet was fed to 10 male and 10 female rats, which means that in total 200 rats were tested. Because only one negative control was used, this control group served as a reference for all nine treatments. As rats, or any other animal, have a serious chance of spontaneously developing different pathologies with age, this one negative control is far too little. On the basis of simple probability calculations, it is already obvious that the chances of finding effects in the group of treated animals are much higher than finding effects in the control group. There should at least have been one control per treatment group. This illustrates the first fundamental flaw of the study: there are too little negative controls.

For their study, Séralini et al. used 'Sprague-Dawley' rats. This laboratory strain has been repeatedly used to test the safety of chemical substances in 90-day feeding studies (e.g. Bondy et al., 2004; Chen et al., 2011). The same rats have also been used in numerous food safety studies using various genetically modified crops over a period of 90 days (e.g. Appenzeller et al., 2008, 2009; He et al., 2009; MacKenzie et al., 2007; Malley et al., 2007). Guidelines that need to be followed for 90-day feeding studies state that at least 10 rats per sex for each treatment should be used (OECD guidelines for the testing of chemicals; health effects; test No. 408; Repeated Dose 90-day Oral Toxicity Study in Rats). However, it is known that from 90 days on, Sprague-Dawley rats have a high propensity to spontaneously develop tumors (Davis et al., 1956; Kaspareit and Rittinghausen, 1999; Suzuki et al., 1979; Schardein et al., 1968; Dinse et al., 2010; Brix et al., 2005) and that the incidence of tumors increases with the age of the rats (Durbin et al., 1966). Although there is nothing wrong with using the Sprague-Dawley rats in two-year feeding trials, their use has consequences for the experimental design. The design should be such that one can distinguish between a tumor that has emerged spontaneously, and a tumor that is the result of the test treatment. National and international guidelines for two-year carcinogenicity studies in rats clearly indicate that each dose group should contain at least 50 animals of each sex (OECD guidelines for the testing of chemicals No. 451; US EPA, OPPTS 870.4200 Health effects test guidelines). With only 10 animals per group per sex the experimental design of Séralini et al. is undoubtedly insufficient. Moreover, when a laboratory does not have an internal standard or historical data of how many spontaneous tumors can develop in their specific lab circumstances, it is crucial to include far more control animals to serve as an internal standard (EFSA, 2011). It is known that the incidence of spontaneous tumors is strongly influenced by the experimental conditions, by the dissection and preparation of tissues for histopathology and by the criteria that are used for the recognition and categorization of tumors. For a good internal standard the number of control animals should be twice as big as the number of treated animals. These are the second and third fundamental flaws in the experimental design: there are far too few animals per dose tested, and there is no correct internal standard to calibrate the number of spontaneous tumors.

Throughout their manuscript, Séralini et al. ignore clear indications that there is something fundamentally wrong in their experimental design. For instance Fig. 1 of the paper shows that among the male animals, there were fewer deaths in the groups that were fed with 22% or 33% genetically modified maize compared to the control. Similarly, there were more deaths among the male rats that had drunk pure water than among those that drunk the highest concentration of Roundup (2.25 g/l). Also Tables 2 and 3 show that the groups that drunk the lowest concentration of Roundup (50 ng/l) were more affected then groups that drunk the highest dose of Roundup (2.25 g/l). In Table 3, it can be observed that the females fed with 33% GMO were much more affected than the females fed with 33% GMO + Roundup. It is clear that the Séralini et al. data are contradictory and do not show any dose–response behavior. Séralini et al. try to explain the absence of a dose–effect relationship by pointing to the fact that hormonal diseases are not proportional to the dose, are non-monotonic and have a threshold effect. But this argument cannot be used if the observed effects are also present in the control group.

From an ethical point of view we have difficulties with the fact that Séralini et al. only show pictures of treated rats that have developed tumors. A picture of a rat from the control group is missing. Similarly, the detailed reporting of the pathologies is strongly biased. From the control groups rats were selected for the absence of tumors, while from the treated groups rats were selected with tumors, suggesting that the observed pathologies were absent in the control group, which is not the case. We also object to the use of the 'two class discriminant analysis' which serves to find differences in general instead of investigating the differences between the treated animals and the control group. A statistical analysis according to accepted standards is missing.

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In summary, we conclude that there are fundamental flaws in the design, analysis and reporting of the Séralini et al. (2012) study, which make it impossible to draw any conclusion. This study should not have been accepted for publication in a peer-reviewed scientific journal. The public debate about the health and safety of genetically modified crops is very sensitive and it is our opinion that the public has the right to be provided with correct scientific information and should not be submitted to unsubstantiated fear.

Conflict of Interest

The authors declare to have no interest in NK603 maize or in Roundup. However, VIB is a world authority in plant research that uses genetically modified plants as a research resource. New knowledge that VIB gathers in this way can, in some cases, contribute to the development of genetically modified crops. For this reason, VIB considers it its social and scientific duty to thoroughly examine new information about the possible health effects of genetically modified plants.

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