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Review

GM crops and the rat digestive tract: A critical review

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ABSTRACT

The aim of this review is to examine the relationship between genetically modified (GM) crops and health, based on histopathological investigations of the digestive tract in rats. We reviewed published long-term feeding studies of crops containing one or more of three specific traits: herbicide tolerance via the *EPSPS* gene and insect resistance via *cry1Ab* or *cry3Bb1* genes. These genes are commonly found in commercialised GM crops. Our search found 21 studies for nine (19%) out of the 47 crops approved for human and/or animal consumption. We could find no studies on the other 38 (81%) approved crops. Fourteen out of the 21 studies (67%) were general health assessments of the GM crop on rat health. Most of these studies (76%) were performed after the crop had been approved for human and/or animal consumption, with half of these being published at least nine years after approval. Our review also discovered an inconsistency in methodology and a lack of defined criteria for outcomes that would be considered toxicologically or pathologically significant. In addition, there was a lack of transparency in the methods and results, which made comparisons between the studies difficult. The evidence reviewed here demonstrates an incomplete picture regarding the toxicity (and safety) of GM products consumed by humans and animals. Therefore, each GM product should be assessed on merit, with appropriate studies performed to indicate the level of safety associated with them. Detailed guidelines should be developed which will allow for the generation of comparable and reproducible studies. This will establish a foundation for evidence-based guidelines, to better determine if GM food is safe for human and animal consumption.

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Abbreviations: EFSA, European Food Safety Agency; FAO, Food and Agricultural Organisation; FSANZ, Food Standards Australia New Zealand; GI, gastrointestinal; GM, genetically modified; H&E, haematoxylin and eosin; LM, light microscopy; OECD, Organisation for Economic Cooperation and Development; RR, Roundup Ready; TEM, transmission electron microscopy; WHO, World Health Organisation.

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1. Introduction

1.1. Background

Genetically modified (GM) or transgenic crops have been grown for human and animal consumption since the 1990s (Clive and Krattiger, 1996). There are currently over 200 different GM crops with various traits approved for human and animal consumption in many countries (ISAAA, 2013). Despite this, feeding studies examining the effects of GM crops on animal and human health are relatively scarce (Domingo, 2000; Domingo and Bordonaba, 2011; Snell et al., 2012).

1.2. Unintended effects and the need for animal feeding studies

The two most common methods of producing GM crops are through *Agrobacterium*-mediated transformation and microparticle bombardment (also known as microparticle acceleration or biolistics) (Wilson et al., 2006). A common criticism is that these processes are imprecise. In both processes, the insertion site of the new DNA is random (Altpeter et al., 2005; Wilson et al., 2006) and more than one copy of the DNA fragment may be inserted into the target genome (Christou, 1992; Gasson, 2003). This can affect gene expression in a positive or negative manner, for example, by causing gene suppression or gene silencing (Altpeter et al., 2005; Dai et al., 2001). In microparticle bombardment, the extra copies of the inserted DNA can be scrambled, inverted or incomplete (Altpeter et al., 2005). In addition, in microparticle bombardment, the site of insertion may undergo further recombination (Altpeter et al., 2005; Christou et al., 1988; Windels et al., 2001). For these reasons, the toxicity or nutritional value of the GM crop should be assessed as a whole.

Transgenic crops are produced through the insertion of a gene cassette, which consists of the desired trait genes, as well as several other genes such as viral promoter and marker genes. These genes tend to be truncated or shortened versions, which may even have gene sequence changes (ISAAA, 2013; Padgette et al., 1995; Vaeck et al., 1987). The effect of these genes acting together is not often determined or even required (FAO/WHO, 2000; FSANZ, 2007).

At present, establishing substantial equivalence is the only generally required safety assessment (FAO/WHO, 2000; FSANZ, 2007). Substantial equivalence relies on the premise that the safety of GM food can be assessed through a comparison with compounds or organisms of known safety. The purpose of the test for substantial equivalence is to identify possible hazard areas, which become the focus of further assessment (FSANZ, 2007; König et al., 2004). The test for substantial equivalence examines the individual characters and not the GM crop as a whole. For example, it assesses the toxicity of the new protein the plant has been designed to produce, such as an insecticidal protein or a protein conferring herbicide tolerance. Based on the safe history of consumption of that protein in its wild-type form, the protein is deemed safe (Kuiper et al., 2001). If the test for substantial equivalence shows no differences outside what could be obtained through natural variation, then food regulators may not require further examinations (Schilter and Constable, 2002). This type of general safety assessment does not consider that the genes present in the novel food may be additional or different from what is anticipated (Padgette et al., 1995; Vaeck et al., 1987; Wilson et al., 2006). It does not take into account the alteration of the protein gene sequence prior to insertion or the possibility that the protein gene sequence may have been altered due to the transformation process, although the latter has recently been incorporated into the European Food Safety Agency (EFSA) assessment processes (EFSA, 2008). Hence, we argue that GM crops should undergo thorough safety evaluations that do not simply consider the GM food as being composed of several substances of known safety, but as a novel entity, the safety of which needs to be evaluated as a whole.

Double- or multi-trait stacked crops are becoming more and more common (Clive, 2013). These are obtained either through more than one trait being inserted into one crop, or through cross-breeding of two or more GM crops (ISAAA, 2013). Many food regulators do not require any studies to be done on crops containing several stacked genes if all the genes in the stack have previously been individually approved for use in the same kind of plant (EFSA, 2010; FSANZ, 2010). However, the effect of two or more traits acting together is unknown. For example, two insecticidal proteins, when ingested together, may have a potentiating or synergistic effect (Schnepf et al., 1998). In real-life scenarios, animals and humans most probably consume GM material and products of various traits in a single meal. Therefore, it is suggested that long-term animal feeding studies be performed to investigate the toxicity of crops possessing more than one trait to investigate the toxicity of feed containing more than one GM component.

1.3. The importance of studying the gastrointestinal tract

The digestive tract is the first site of contact for any ingested compound. It follows that if a compound is toxic, the first signs of toxicity may be visible in the gastrointestinal (GI) tract. Furthermore, since the stomach and the intestines are the sites of longest residence for any ingested product, these should become the most important sites for the evaluation of an ingested compound's toxicity. It is difficult to assess damage to the digestive tract purely on macroscopic grounds (Morini and Grandi, 2010), therefore a histopathological analysis should be part of the investigation.

2. Methods

The purpose of this literature review was to examine the relationship between GM crops and histopathological observations in rats. The search only included crops possessing one or more of three specific traits which are commonly found in commercialised GM crops: herbicide tolerance via the *EPSPS* gene, and insect resistance via *cry1Ab* or *cry3Bb1* genes. A list of crop event names was first generated (Table 1) based on GM approval databases (CERA, 2012; FSANZ, 2011b; ISAAA, 2013) and publications, such as literature reviews (Domingo, 2007; Domingo and Bordonaba, 2011; Magaña-Gómez and De La Barca, 2009; Pusztai et al., 2003; Snell et al., 2012). The search used PubMed, Google Scholar and Embase to find studies that were published before April 2013. The search was restricted to published studies. Reports, such as EFSA reports, were not included since they do not contain detailed histopathological results. The keywords used were rat, rats, *rattus* and the specific crop event line name (Table 1). To make results comparable with each other, the search was limited to long-term rat feeding studies of no less than 90 days duration. The search excluded multigenerational studies, unless there was a histopathological investigation in the first generation of rats. No language limit was set. For non-English publications, help was obtained with their translation and accurate understanding.

3. Results

The search yielded 21 published studies (Table 2) with an additional two re-analyses of raw data of some of these studies (de Vendomois et al., 2009; Seralini et al., 2007). The re-analyses concentrated only on the blood, serum and urine test results. (These publications are not counted nor listed in the tables or figures since they are not original feeding studies). Eighteen (86%) out of the 21 studies investigated crops that have been approved for human and/or animal consumption somewhere in the world (Table 1). These 18 studies investigated only nine out of the 47 approved GM crops (19%) known to possess at least one of the traits of interest. No published rat-feeding studies could be found for the remaining 38 (81%) approved crops. Of all the 21 studies found, 12 (57%) generally assessed the long-term effect of GM feed on rat health (Hammond et al., 2004, 2006a,b; Healy et al., 2008; Qi et al., 2012; Sakamoto et al., 2007, 2008; Schrøder et al., 2007; Seralini et al., 2012; Tutel'ian et al., 2008, 2010; Wang et al., 2002), whilst seven