sustainability

MDPI

Perspective

A Snapshot on Food Allergies: A Case Study on Edible Flowers

Massimo Lucarini [1,*], Andrea Copetta [2], Alessandra Durazzo [1], Paolo Gabrielli [1], Ginevra Lombardi-Boccia [1], Elisabetta Lupotto [1], Antonello Santini [3] and Barbara Ruffoni [2,*]

[1] CREA-Research Centre for Food and Nutrition, Via Ardeatina 546, 00178 Rome, Italy; alessandra.durazzo@crea.gov.it (A.D.); paolo.gabrielli@crea.gov.it (P.G.); g.lombardiBoccia@crea.gov.it (G.L.-B.); elisabetta.lupotto@crea.gov.it (E.L.)

[2] CREA-Research Centre for Vegetable and Ornamental Crops, Corso Inglesi 508, 18038 Sanremo, IM, Italy; andrea.copetta@crea.gov.it

[3] Department of Pharmacy, University of Napoli Federico II, Via D. Montesano 49, 80131 Napoli, Italy; asantini@unina.it

* Correspondence: massimo.lucarini@crea.gov.it (M.L.); Barbara.ruffoni@crea.gov.it (B.R.)

Received: 16 September 2020; Accepted: 13 October 2020; Published: 20 October 2020

Abstract: This perspective study addresses the main causes of adverse reactions to foods in humans, by taking into account the main allergic reactions that may occur as a result of food ingestion, as well the main allergens present in food and how their allergenicity change as a result of food preparation. In addition, European legislation on food labeling and novel foods was taken into account. The case study of this perspective is on the potential allergenicity of edible flowers as well as evidence of phytochemistry and toxic compounds and the risk associated with their ingestion. Regarding edible flowers, a key issue to address is if they are safe to consume or not. In the framework of the project "Innovative activities for the development of the cross-border supply chain of the edible flower" (ANTEA), we considered 62 different species and varieties of edible flowers. The results obtained by consulting two databases on allergens, COMPRISE and Allergen Nomenclature, marked two alerts for two species of edible flowers selected in the project. Moreover, based on edible flower consumption, about ten grams per serving, and on their protein content, we can also state that the risk of allergic reactions due to edible flower ingestion is very low.

Keywords: food allergy; allergens; allergic symptoms and syndromes; cross-contamination; cross-reactivity; safety; edible flowers; ANTEA project

1. Food Allergies

Adverse reactions in humans due to food ingestion can produce different and sometimes even very serious clinical conditions. Body reaction to food intake may produce toxic effects, and have an impact on sensitive individuals. On the other hand, body reactions to food depend mostly on the susceptibility of each individual to a particular food, while the same foods are innocuous to a non-sensitive individual. This particular reaction to food is referred to as an allergy and is sustained by an immunological mechanism. The allergic reactions are quite often due to immunoglobulin E (IgE). If no immunological mechanism is involved in the adverse reaction, the reaction is defined as an intolerance. According to the mechanism involved, Figure 1 reports a scheme on adverse reactions.

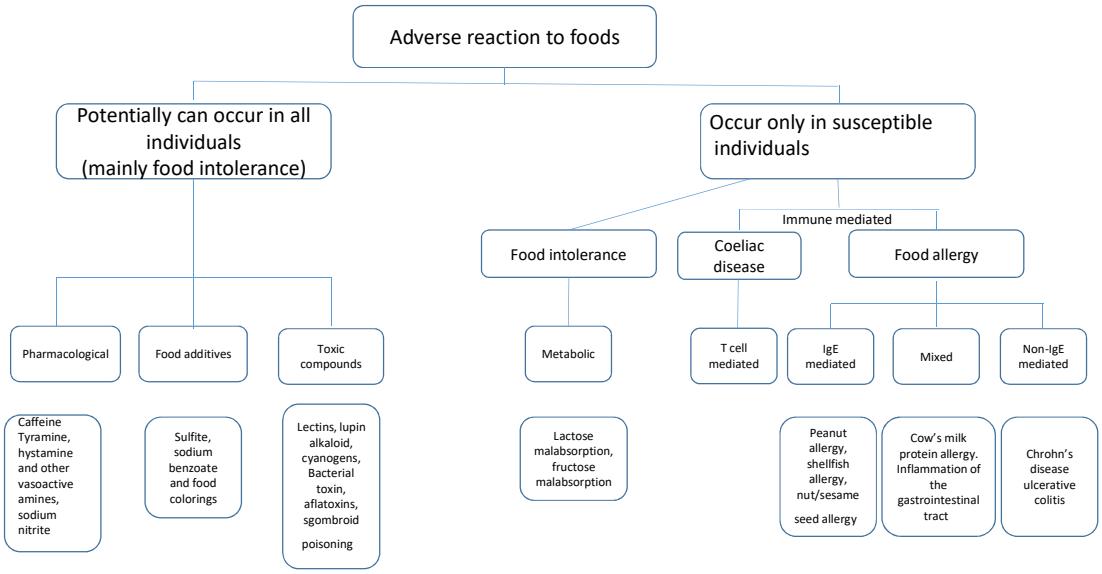


Figure 1. Overall Scheme of Possible Adverse Reactions to Foods.

As shown in Figure 1, adverse reactions to foods can be grouped into two main categories: the ones which can occur in all individuals who consume the food (food intolerances) and those that occur only in those individuals who are defined as sensitive. Reactions that do not involve the immune system are due to foods that may contain toxins, pharmacologically active compounds, or those affected by microbiological contamination. Therefore, food intolerance can refer to (i) an adverse physiologic response to intrinsic food properties (i.e., pharmacologic active substances and/or toxic contaminants) or (ii) the host characteristics (i.e., metabolic disorders), often not reproducible, that are often in a major part of cases dose dependent [1].

These reactions can be developed by all individuals if the intake of the contaminant is in an amount capable to trigger the unwanted effect in the body. Some of these food intolerance reactions can mimic the effects of an allergic reaction. For example, scombroid syndrome is a pseudo-allergic poisoning caused by the consumption of inadequately preserved fish. It is among the most common types of fish poisoning in the world, since this species of fish is consumed in large quantities. Such a reaction affects any individual who eats an amount of the fish capable to stimulate an allergic-like reaction, but it is not linked to any real allergic mechanisms.

Therefore, the term food hypersensitivity is used preferentially when referring to food intolerance with the aim to define any abnormal, reproducible, non-psychologically mediated reaction to food consumption.

Sometimes the mechanism of food intolerance is known (e.g., involves an enzyme deficiency or the presence of substances with pharmacological effect) but, in a major part of cases, the mechanism of the hypersensitivity is unknown.

On the other hand, the food allergy [2–4] is referred to as an abnormal immunologic response to a food occurring in a susceptible individual and can be classified as IgE mediated (when the immune system is responsible for the food allergy), cell mediated (when the interactions between cells and chemical mediators are responsible for the food allergy) , and mixed IgE–cell mediated when both IgE and immune cells are involved in the allergy mechanism (see Figure 1).

The allergic reactions are reproducible, and they are often not dose dependent. The most common food allergies have been observed for foodstuffs like eggs, milk, peanuts, wheat, soy, tree nuts, fish, and shellfish (all included in the allergenic foods category).

Another mechanism can take place later in life in patients suffering from respiratory allergies to pollen who represent another large class of sensitive individuals with a food allergy to vegetables and fruits. This allergic reaction called oral allergic syndrome (OAS) occurs in the oral mucosa.

Those individuals who are allergic to pollen sometimes have to be also careful of what they eat. A simple apple, a peach, or an inviting strawberry can be dangerous for those who are allergic to birch, just as tomatoes, potatoes, and citrus fruits can irritate the lips and palate of those who are allergic to gramineous plants. These are the so-called cross-allergies, a disorder caused by a cross-reaction between pollens from some families, and different kinds of foods.

This phenomenon occurs because many foods contain protein molecules structurally similar to the ones which can be found in pollen. These molecules are recognized by the immune system, this way triggering the cross-food reaction onset. In these cases, those individuals who are allergic to pollen produce IgE immunoglobulins to specific food proteins that have a molecular structure highly homologous to that of respiratory allergens (cross-reaction): the first step is the sensitization to pollen and then the sensitization to the cross-reactive food allergen. The prevalence of OAS ranges from 6% to 47% and occurs in 47% to 70% of patients suffering from pollen allergy, in relation to the type of pollen to which the individual is exposed [5].

Nonetheless, it is important to emphasize that not all people allergic to pollen necessarily have a cross-allergy to food, and also that not all the foods reported in Table 1 can give allergies to a single person. For these reasons, it is important that only a specialist is in charge to indicate for each patient whether to follow a specific diet and which diet to follow in order to limit the risk of nutritional imbalances. The perceptions of adverse reactions to food by the population are common. Many studies have shown that although about 20%–30% of people believed that their allergic reaction symptoms were food consumption related, after double-blind placebo-controlled food challenges, only the 2%–3% of the subjects showed a positive adverse reaction to food. The prevalence of food allergy in the general population has been estimated between 2%–4%. In children, the prevalence is around 6%–8%, and the allergy to eggs and cow's milk are the most important forms, while in adults the higher prevalence of food allergies is shown towards shellfish and peanuts. This difference in sensitization to food proteins among adults and infants can be attributed to incomplete development, for the latter, of the gastrointestinal immune system which is still immature and the intestinal physiology is not complete (for example, in terms of permeability) and sensitization of IgE to food proteins may occur. Individuals usually normally develop a tolerance to ingested food proteins, which are harmless at an early age, and the prevalence of food allergies slightly decreases with age [6].

1.1. Mechanisms of Food Allergy

Food allergy is an immunological reaction versus a food antigen that occurs soon after eating food. As reported in Figure 1, the mechanisms involved in a food allergy can be IgE mediated, non-IgE mediated, or mixed (IgE and non-IgE mediated).

Almost all food allergies concern the production of immunoglobulin E (IgE) and the development of interactions between different typologies of cells and chemical mediators. Immediate symptoms are produced by this type of reaction (Type I reaction), and among all symptoms, anaphylaxis is the most severe form [7–10].

Generally, allergic reactions are mainly caused by a few foods (eggs, milk, tree nuts, peanuts, fish, shellfish, wheat, and soy) that have common characteristics: they are glycoproteins, soluble in water and stable to heat, acids, and digestive enzymes, with a molecular weight between 10 and 70 kD. Another recognized mechanism in food allergies is an allergy that is not mediated by IgE (Type IV reaction), which develops after some hours or days after exposure (the so-called delayed cell-mediated allergy) [11]. They are less characterized than the IgE mediated but are typically due to chronic inflammation. In this case, the key mechanisms are the interactions between cells and chemical mediators, in place of immunoglobulines. Type IV reactions can likely occur in response to the ingestion of several foods and can lead to symptoms in different parts of the body such as the skin, intestine, and other organs.

Regarding the immediate IgE-mediated reactions, the antibodies are proteins produced by the B lymphocytes in response to the presence of body-foreign proteins [12]. The biological abilities of antibodies are to counteract infection with bacteria, protozoa, and viruses. The body maintains the

function to produce immunoglobulines against an individual allergen along with time and sometimes for life. The allergy response degree can be different in relation to the specific antigen and taking into account the levels of exposure.

Many substances that may be ingested, inhaled, or come into contact with the body (i.e., pollens, house-dust mites, molds, and foods) can contain potential allergens. This can give rise to the unwanted or excessive formation of antibodies in individuals that are susceptible. The five classes of immunoglobulin are IgA, IgD, IgE, IgG, and IgM.

The IgE produced after exposure to an allergen acts as a bridge among the antigen and surface of immune cells such as mast cells and, thus, the mast cells become sensitized to the specific allergen.

If the sensitized mast cells are re-exposed to the same antigen (or could be enough to the same epitope in a different antigen), an IgE-mediated allergic response occurs [10,13,14] and several chemical mediators (i.e., histamine) are released; in the chain reaction, other pro-inflammatory substances, i.e., leukotrienes and prostaglandins, can be released; this can lead to an immediate inflammatory response, which results in local swelling, itching, redness, and heat.

In the immediate response, mediators are released by mast cells and T lymphocytes, and more IgEs are produced by B lymphocytes [15]. The inflammatory response is intensified by a range of chemicals (e.g., toxic proteins) released by eosinophils activated by mediators from T lymphocytes.

Mast cells and basophils are triggered after encountering the food allergen, producing local and a widespread reaction. In the case of the non-allergenic process, foods containing substances that can trigger mast cells to release histamine and other mediators (pineapple, strawberries, egg white, and/or other foods or food additives) could also give rise to the same results of an activated mast cell, thus, mimicking an IgE-mediated allergic reaction.

1.2. Sensitization and Cross-Reactivity

Even if many potentially allergenic substances can be present in food, allergy due to inhaled substances seems to be more common than food allergy because the intestine gives a series of defensive barriers, thus, avoiding contact among food allergens and mast cells in the gut wall [16].

The response provoked by the first exposure to an antigen, throughout the gut or in another location, can give rise to one of the following conditions:

- the individual becomes "tolerant" to the allergen, and subsequent exposures will not result in the production of antibodies;
- the individual becomes "sensitized" to the allergen, and subsequent exposure develops an IgE-mediated response with unwanted symptoms; or
- the individual develops an immune response, involving the production of other types of immunoglobulins (i.e., IgG) that may reappear on consequent exposure, but that does not necessarily give symptoms.

The amount of allergen required for the sensitization is not completely known, and it is difficult to assess a relationship between dose and allergenic effect.

The match of IgE to its antigen is specific: it does not match the entire antigen molecule but only the epitope. Other antigens with a very similar epitope could react with the same IgE, giving rise to a cross-reactivity (also known as cross-allergenicity) reaction; these reactions occur when an individual is exposed to a second antigen after sensitization to a first [17].

Co-sensitization indicates an individual sensitization to more than one allergen source that is not due to cross-reactivity, as it is not mediated by shared epitope-specific antibodies [18,19]. Florin-Dan Popescu [20] well summarized important allergen component families such as serum albumins, lipid transfer proteins, and tropomyosins, which are involved in cross-reactivity between aeroallergens and food allergens [18,20–23].

An example of cross-reactivity is given by the fact that the sensitization to birch pollen can lead to oral allergy to various fruits. Other examples of cross-reactivity are reported in reference [24]. In Table 1, the main cross-reactors are indicated.

Table 1. The Main Cross-Reactors Based on Reference [24].

Environmental Allergen	Fruits	Vegetables	Nuts	Spices	Legumes and Other Foods
Tree pollen	Apple, apricot, cherry, kiwi, nectarine, peach, plum, prune, strawberry, lychee, jackfruit, Sharon fruit, tomato	Carrot, celeriac, celery, green pepper, potato, parsnip	Walnut, almond, hazelnut	Basil, caraway, anise, dill, thyme, pepper, tarragon, paprika, fennel, marjoram, oregano, parsley, cumin, coriander, chicory	Bean, pea, soybean, peanut, lentil, sunflower seed
Grass and grain pollen	Kiwi, orange tomato, melon, watermelon, date	Potato			Peas, peanut, sunflower seed, flour, bran, legume
Mugwort pollen	Mango, grapes, pineapple, avocado, banana, tomato, lychee, peach, watermelon, melon, apple, orange	Celeriac, parsnip, carrot, celery, onion, green pepper, potato	Chestnut	Basil, caraway, anise, dill, thyme, pepper, tarragon, paprika, fennel, marjoram, oregano, parsley, mustard, coriander	Sunflower seed, chamomile
Ragweed	Melon, banana, watermelon	Zucchini, cucumber			

1.3. Adverse Reactions to Foods: Symptoms and Syndromes

An allergic reaction to a single food is simply detected, but the diagnosis of adverse reactions to foods is difficult because other foods can be involved, giving time-delayed adverse reactions and side effects. An individual food can cause different symptoms in different people, or even in the same person at different doses or at different times; the mechanisms involved in these reactions can also vary, as well. Indeed, diagnosis of acute allergic reactions can be evaluated relatively simply by the dietary history of an individual because they usually take place within minutes after the ingestion of the food.

True food allergy often may give rise to a huge spectrum of symptoms, influencing many parts of the body as reported in Table 2.

Table 2. Possible Symptoms Due to Food Allergy Based on References [25,26].

Symptoms Which May Result from Food Allergy	IgE Mediated (Immediate Reaction)	Non-IgE Mediated (Delayed Reaction)
Systemic		
Anaphylaxis	✓	
Gastrointestinal		
Swelling and itching of lips and mouth	✓	
Nausea	✓	
Vomiting	✓	✓
Diarrhea	✓	✓
Crampy pain	✓	✓
Respiratory		
Rhinitis	✓	
Asthma	✓	
Swelling of larynx	✓	
Cough	✓	
Chest tightness	✓	
Bronchospasm	✓	
Skin		
Pruritus	✓	✓

Urticaria	✓	
Erythema	✓	✓
Eczema	✓	✓
Conjunctivitis	✓	
Cardiovascular		
Presyncope/syncope	✓	
Hypotension	✓	
Tachycardia	✓	

2. Food Allergens

2.1. Classification of Food Allergens

Chemical structures (sequence and three-dimensional feature) of proteins play a key role in their allergenic capacity; in fact, food allergens appear to be limited to only a few protein families. Hoffmann-Sommergruber and Mills [27] reviewed the protein families on the basis of their structural characteristics and their potential allergenicity within the EuroPrevall project [27].

The main proteins involved have specific biological activities (i.e., hydrolysis of polysaccharides and proteins, binding of lipids and ions, storage and transport, and cytoskeleton organization) [28]. In Figure 2, the main protein families with allergenic activity and food sources are shown.

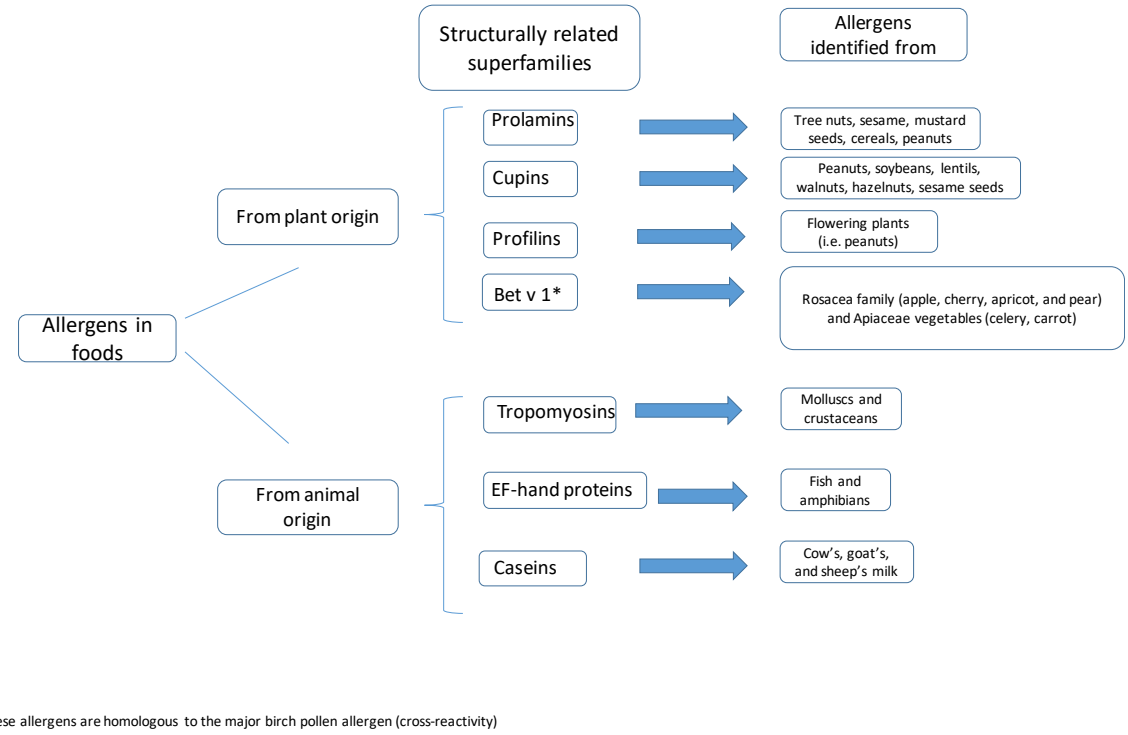


Figure 2. A Shot of the Main Food Allergen Protein Families' Distribution [27–31].

It is noteworthy that, although the common structural characteristics of proteins and biological functions have been provisionally due to their immunogenicity, the prediction of the allergenicity of a protein cannot be based only on these two parameters [32]. As reported by Dall'Antonia et al. [33], promising tools have been studied on the prediction of structure protein.

Concerning the threshold levels of allergen [34], the sensitivity to single food allergens is different among people, and maybe also for the same person at different times; due to this variability, the lowest dose of a food allergen that can stimulate an adverse reaction is difficult to predict. For instance, Blom et al. [35] reported the threshold dose for five main allergenic foods in children; the results of reference [35] showed that the protein dose at which 5% of the allergic

population is likely to respond with objective reactions was 1.1 mg for cow's milk, 0.29 mg for hazelnut, 1.5 mg for hen's egg, 1.6 mg for peanut, and 7.4 mg for cashew nut. The same authors also remarked how threshold distribution curves allow the comparison of various allergenic foods and to inform on precautionary labeling.

Nowadays, across Europe, fourteen groups of foods are identified as the most common ingredients or processing aids leading to food allergies and intolerances [36] (see paragraph 4).

2.2. Database of Allergens

The database Allergen Nomenclature at website reference [37] includes approved and officially recognized allergens. The systematic allergen nomenclature is required by many peer-reviewed scientific journals.

Another example is the COMprehensive Protein Allergen REsource (COMPARE) database [38] that is based on an exhaustive listing of peer-reviewed and clinically relevant protein allergens with citation support and species identification. In the database are the descriptions of the allergens and the amino acid sequences.

Other examples are also described in the work of Tong et al. [39], and further in that of Radauer et al. [40] that gives a picture of main allergen databases and their applications. For instance, Allergome [41] is a comprehensive collection of data on compounds that cause IgE-mediated diseases selected from web-based resources and international scientific journals. AllergenOnline [42] gives access to a peer-reviewed allergen list and sequence searchable database that can allow for the identification of proteins potentially related to allergenic cross-reactivity.

The Structural Database of Allergenic Proteins (SDAP) [43] represents a tool for investigating the cross-reactivity between known allergens, in examining the FAO/WHO allergenicity guidelines for new proteins, and for predicting the IgE-binding potential of genetically modified food proteins. The SDAP browser allows one to retrieve information related to an allergen from the most common protein sequence and structure databasesin order to identify the sequence and structural neighbors of an allergen, and also the presence of an epitope.

The Immune Epitope Database (IEDB) [44] represents a useful tool for the prediction and analysis of epitopes; it can be considered a catalogue of experimental data on antibody and T cell epitopes.

3. Food Processing and Allergenicity

The stability of allergens is defined as the capacity to maintain their configuration after thermal, chemical, or enzymatic (proteases) processing [32,45]. The recent work of Perkar et al. [46] highlights the peculiar characteristics and molecular patterns that contribute to the stability of protein structures and consequently to allergenicity. In the evaluation of the allergenic potential of food proteins, the encountered gastrointestinal conditions, such as food matrix, enzyme levels, pH, and the immune status of the individual, should be considered.

3.1. Influence of Processing on Food Allergenicity

As widely reported in the literature, bioactive compounds content can be significantly influenced by cooking and food processing [47,48]. Foods and food ingredients are subject to different treatments (i.e., cooking) and technological processes (extrusion, boiling, stewing) in order to improve their palatability and favor digestion, as well as to inactivate pathogenic microorganisms and/or eliminate toxins. Food can be processed at home, in restaurants, and by the food industry.

The processing produces physical and chemical changes that lead to alterations of different components, including protein and, if any, its allergenicity. Particularly the epitope present in the food matrix could be destroyed by food processing or new epitopes could be formed [49]. Food-processing techniques can influence the structure and chemical properties of proteins and indeed may alter the allergenic properties of food proteins. Main modifications strictly linked to allergenic potential are protein unfolding and aggregation, glycation and glycosylation, proteolysis,

and the formation of products of the Maillard reaction [50]. The nature of the protein, the process conditions, and the composition of the food matrix are the main influencing factors. Particularly, the types and conditions of the process affect the chemical structure of proteins [51]. Lepski and Brockmeyer [52] well discussed the influence of processing on food allergen structure and consequently on their allergenicity, with particular emphasis on partial loss of conformation, chemical modification, and the impact of novel processing techniques. A systematic investigation data marked how the literature is scarce on the influence of food processing on allergenicity [53].

Verhoeckx et al. [54], by reviewing the influence of food processing (i.e., heating) on the allergenic potential of the common food allergens (hen's eggs, cow's milk, tree nuts, peanuts, soy, mustard, wheat), concluded that processing does not completely inactivate the allergenic activities. Although heating could cause changes in single proteins, they may lead into an increase (i.e., from Maillard reaction products) or a decrease (i.e., in extensively heated egg white) of allergic sensitivity. In particular, the authors reported how only microbial fermentation and enzymatic or acid hydrolysis could reduce allergenic integrity and consequently allergenicity to the point that symptoms will not be exhibited; other methods could be promising but to date, more extensive studies and data availability are needed [54]. Vanga et al. [55] well described and discussed the impacts of conventional and innovative food processing methodologies on the activity of food allergens. It is worth mentioning the recent work of Cabanillas and Novak [56], which gives an updated view, taking into account several variables, of the influence of processing techniques on the allergenicity of different foods.

Processing methods can have a different impact on certain foods with respect to other foods: food processing could decrease, not change, or even increase the allergenic activity [57]. Several authors highlight how it is difficult to predict the multiplicity of allergenic foods/ingredients related to the multiplicity of the allergenic structures in a whole food and that the same treatment could differently influence different proteins. Moreover, it has been also been remarked upon how integrated and robust methods to assess the risk of food allergenicity should be developed [54,58–61].

Several authors have shown how some fruits induce more frequent allergic reactions when consumed with peel [62–64]. Concerning the fruit allergy, it is worth mentioning the critical review of Wang et al. [65] on the influence of pre-harvest and post-harvest conditions on the fruit allergenicity: the environmental and cultivation conditions (nitrogen, water shortage, climate factors, etc.) [66] as well as the variety [67], harvesting maturity [68], and storage [69] can affect the allergenicity potential.

Regarding the thermal processing, one of the main interactions between proteins and sugars that occur in food during heat processing is the Maillard reaction (MR). Teodorowicz et al. [70] and Gupta et al. [71] discussed the pros and cons of the Maillard reaction in food allergies. Toda et al. [72] well discussed how products of Maillard reactions can affect allergic diseases from the effects on the digestibility of proteins to influence on the immunogenicity of proteins.

Gupta et al. [71] discussed the pros and cons of the Maillard reaction in food allergies; particularly, the authors summarized both the studies reporting the impact of the Maillard reaction of enhancing allergenicity in food allergens, i.e., eggs [73], peanut [74], potato [75], and codfish [76], as well as studies reporting the impact of the Maillard reaction with decreased allergenicity in food allergens, i.e., eggs [77,78], peanuts [79], and hazelnut [80].

The work of Verma et al. [81] well described how thermal processing can influence legume allergens and explored the different approaches to reduce or eliminate allergenicity of leguminous food using various thermal processing (i.e., autoclaving, blanching, microwave heating, pasteurization, steaming, or canning) of legumes. These last may reduce, eliminate, or enhance the allergenic potential of a respective legume, but in most of the cases, minimization of allergenic potential was reported. For instance, different studies reported how boiling has little effect on legumes' allergenicity [82–84]; allergic reactions were even reported after the inhalation of boiling vapors in sensitized patients [85].

Cabanillas et al. [58] showed how thermal processing influences the allergenic potential and the IgE cross-reactivity of legumes (particularly soybean and peanuts) with particular regard to boiling, roasting, and frying. Concerning the effect of processing technologies on the allergenic properties of egg proteins, some studies showed that the reduction of the allergenicity is related to the protein conformational changes [86].

Various non-thermal processing technologies (e.g., irradiation, ultrasound, pulsed electric field, cold plasma, and high-pressure treatments) can influence the protein structure, and consequently their solubility and their functional properties [87].

For instance, Huang et al. [88] has reviewed the potential utility of high-pressure processing in reducing food allergenicity. The recent work of Yang et al. [89] showed how solid-state fermentation with microorganisms reduces the allergenicity of soybean meal through degradation of the main allergens.

4. Labeling and Novel Foods: European Regulations

4.1. Labeling

Among the European Directives, the only legal acts that refer to food allergens are the Labelling Directive (Directive 2000/13 / EC) [90] and its subsequent amendments. Following the Labelling Directive, the EU requires manufacturers to include the ingredients present on all prepackaged food products in order to be sold with very few exceptions. Several amendments have been tabled and accepted to modify this directive. The two most important amendments are as follows.

(1) Directive 2003/89/EC [36] (and further amendments) introduced Annex IIIa, which is the list of fourteen allergenic foods that, if present in a product, must always be shown on the package label, as follows:

◎Cereals containing gluten (i.e., wheat, rye, barley, oats, spelt, kamut, or their hybridized strains) and products thereof

Crustaceans and products thereof

Eggs and products thereof

Fish and products thereof

Peanuts and products thereof

Soybeans and products thereof

Milk and products thereof (including lactose)

Nuts i.e., almonds, hazelnuts, walnuts, cashews, pecan nuts, Brazil nuts, pistachio nuts, macadamia nuts, and Queensland nuts, and products thereof

Celery and products thereof

Mustard and products thereof

Sesame seeds and products thereof

Sulfur dioxide and sulfites at concentrations of more than 10 mg/kg or 10 mg/liter expressed as SO_2.

Lupin and products thereof

Mollusks and products thereof

(2) Directive 2007/68/EC [91] is the most recent amendment of Annex IIIa. It contains the list of allergens that must be labeled and the products deriving from these allergens for which it is not necessary to add the allergen on the label.

The web site of the European Food Safety Authority (EFSA) offers guidance on the labeling of food allergens for Europe. When allergens are deliberately added to a food as ingredients, Regulation (EU) no. 1169/2011 [92] requires the indication of allergens on both prepacked and non-prepacked foods. The quantity of allergens that are intentionally added to a food is not subject to any threshold with the sole exception of sulfites and sulfur dioxide. The presence of traces of an allergen used as an ingredient in a food product must be reported on the label (https://ec.europa.eu/food/sites/food/files/safety/docs/codex_ccfl_cl-2018-24_ann-02.pdf). The labeling workflow is simplified in Figure 3.

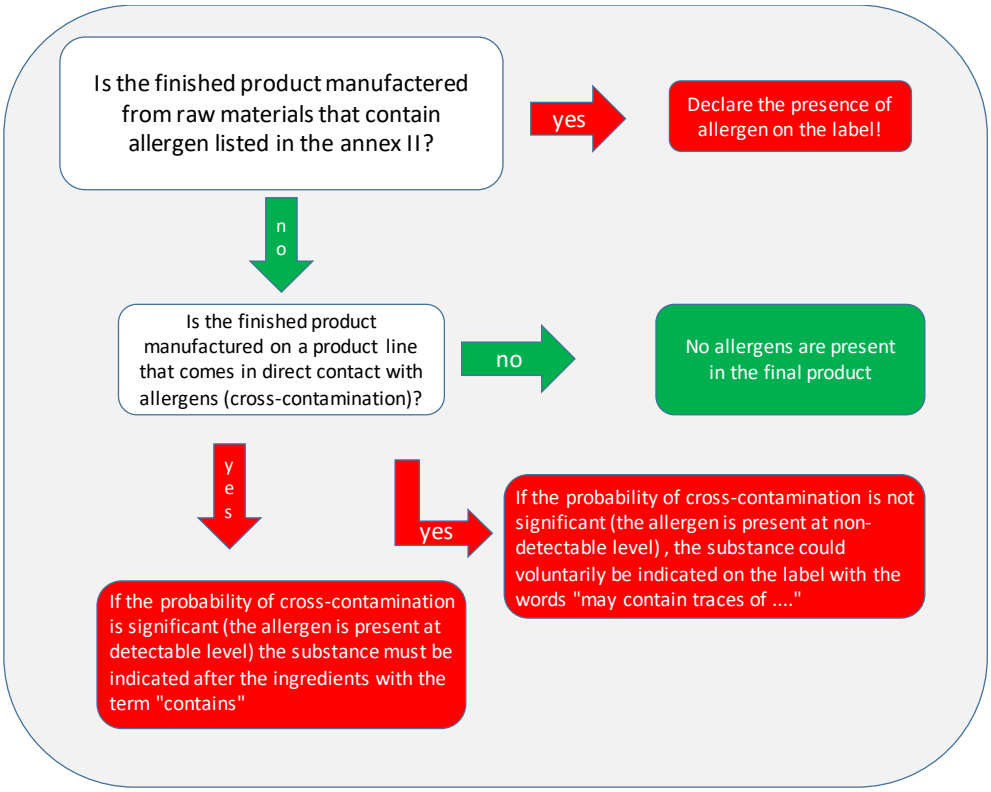


Figure 3. Description of labeling workflow.

As shown in Figure 3, allergens could enter in the food production chain accidentally through, for example, the use of common processing equipment or too close production lines. The Codex recommendations or the EU Labelling Directive do not cover this cross-contact. Some European countries, i.e., the UK and Italy, have created national guidelines to aid the food industry in how to manage and label allergens including cross-contact allergens (Figure 3).

4.2. Novel Food Regulation

As indicated by EU regulations, novel food is a term which defines and can be used for all the food which were not consumed by humans as part of the diet before 15 May 1997 [93] (Regulation EC No. 258/97 of the European Parliament and of the Council of 27 January 1997 concerning novel foods and novel food ingredients, *Official Journal of the European Comm.* L43, 40, 1997. ISSN 0378-6978).

The following must be considered novel food: (i) newly developed foodstuff; (ii) innovative food; (iii) food produced applying new processes or new technologies; (iv) food currently or historically consumed outside the European Union. There are many examples regarding the novel food concept, e.g., new vitamin K sources (menaquinone) or extracts from existing food (Antarctic krill oil rich in phospholipids from *Euphausia superba*);third countries' crops such as noni fruit juice and chia seeds; food produced applying new processes or new technologies like UV-treated food (bread, milk, mushrooms, and yeast).

The main issues to be considered on novel food are safety for consumers, proper labeling, and not replacing a normally used food or, if this is the case, not altering the nutritional value of the replaced food. For these reasons, novel foods require a pre-market authorization and must follow the current regulation valid since 1 January 2018—Regulation (EU) 2015/2283 [94] on novel foods, replacing Regulation (EC) No 258/97 and Regulation (EC) No 1852/2001 which were in force until 31 December 2017 (https://ec.europa.eu/food/safety/novel_food/legislation_en).

This new regulation expands categories of novel foods, and defines the various situations of foods originating from plants, animals, microorganisms, cell cultures, minerals, etc., specific categories of foods (insects, vitamins, minerals, food supplements, etc.), foods resulting from production processes and practices, and state of the art technologies (e.g. intentionally modified or new molecular structure, nanomaterials), which were not produced or used before 1997 and thus may be considered to be as novel foods. Under the new Regulation, all authorisations (new and old) are generic as opposed to the applicant-specific, restricted novel food authorisations under the old Novel Food regime.

This means that as long as the authorized conditions of use, labeling requirements, and specifications are respected, an authorized novel food can be placed on the European Union market by any food business operator.

A list containing all authorized novel foods has been set up (https://ec.europa.eu/food/safety/novel_food/authorisations/union-list-novel-foods_en) [95].

Future authorized novel foods will be included in the Union list with Commission Implementing Regulations, by means of simplified procedures controlled by the European Commission. Safety issues will be addressed by the EFSA (European Food Safety Authority), which will follow efficiency and transparency criteria, and will shorten the overall time on approvals by setting up deadlines for safety evaluation and authorization procedures. A new aspect is also the possibility to fast evaluate the safe use of traditional food from third countries, and to provide simplified guidelines to evaluate foods new to the European countries. Therefore, the traditional food will be released on the European Union market provided that no safety-related objections are raised by the EFSA or the European countries. An applicant can obtain a 5 year-limited authorization for placing a novel food on the European market by providing proprietary and scientific safety evidence. To easily assess whether an authorization is required under the Novel Food Regulation, a non-exhaustive catalogue of novel foods has been developed (https://ec.europa.eu/food/safety/novel_food/catalogue/search/public/index.cfm) [96].

5. Focus on the Edible Flowers

5.1. Edible Flowers: A Source of Bioactive Compounds

Cunningham in reference [97] well represented a picture of main topics regarding edible flowers which are nowadays becoming the latest food fashion, challenging this niche market. Many ornamental flowers and wildflowers can be employed to garnish dishes and as ingredients in recipes.

Edible flowers are harmless and non-toxic flowers that contain healthy compounds useful in the human diet; producers and merchants of edible flowers are actually increasing. The culinary use of flowers as ingredients in food preparations, on the other hand, is historically documented, and it is treated differently according to cultural environments [98–100].

Flowers were chosen for culinary preparations in Greece, Egypt, and in Roman times; the first records on the use of flowers date back to 140 BC [101]. Native flower species are used worldwide in food preparations, and are safely eaten, or used for garnishing dishes and drinks.

People looking for food innovative propositions, cooking or garnishing dishes with flowers, is gaining increasing interest. In this order, new knowledge about the presence of bioactive compounds and the nutritional value of edible flowers is needed. [102]. Recently, Fernandes et al. [103] gave a picture of the edible flower market. This aspect triggers also the need for more in-depth assessment of legislation and safety regulation to better assess the future trends of edible flowers.

Rop et al. [104], by studying the nutritional composition of 12 species of edible flowers, reported higher mineral content in chrysanthemums, dianthus flowers, and violas. The most abundant element was potassium. Among the ingredients of traditional Japanese cuisine, there are also flowers; therefore, Chemson et al. [105] studied compounds with nutritional and nutraceutical character in thirteen edible flowers.

As reported for vegetables and fruits, the colors of flowers suggest the presence of phytochemicals [106–110]. A recent work by Pires et al. [111] presents the phenolic profile of several samples of *Calendula officinalis*, *Centaurea cyanus*, dahlia mignon, and various rose species (Rosa damascena 'Alexandria' and *Rosa gallica* 'Francesa' draft in *Rosa canina*), and revealed their biological potential. Another current example was given by Nowicka and Wojdyło [112] that reported the beneficial anti-hyperglycemic and anticholinergic capacities deriving from the presence of natural antioxidant compounds in tissues of edible flowers such as hawthorn, primrose, and linden blossom.

Various processing and conservation technologies have been tested to extend the shelf lives of edible flowers and, thus, ensure their commercialization; in this regard, Zhao et al. [113] summarized the innovative and emerging technologies (i.e., high hydrostatic pressure, modified atmosphere packaging, microwave drying, irradiation, freeze drying, and hybrid drying) to preserve the flowers in excellent condition, as well as different extraction techniques to obtain and microencapsulate the phytochemicals.

The US Department of Agriculture Nutrient Database reported the nutrient composition of certain edible flowers, such as broccoli, various types of courgette blossoms, hibiscus flowers, and white-flowered calabash, and some derivatives such as wild flower honey. [114]. Another example is reported in the Bioactive Substances in Food Information Systems (eBASIS) Database [115] (http://ebasis.eurofir.org/Default.asp) where the profiles of bioactive compounds of some edible flowers are described, such as red clover, rowan, purple coneflower, perforate and common St. John's wort, pale Echinacea, marigold, lemon balm, cauliflower, chamomile, broccoli, and artichoke.

5.2. Safety Issues of Edible Flowers

Edible flowers are defined as nontoxic, innocuous flowers with health benefits [116], but from a nutritional point of view, the benefits and risks related to their consumption must be considered. In fact, if it is true that flowers are an important source of nutrients and molecules with high antioxidant activity, it is also necessary to consider another topic to be addressed scientifically related to their safety of use for the possible presence of unwanted substances in and/or on flowers.

From the point of view of adverse reactions of edible flowers consumption, one should consider the following:

- the presence of toxic compounds and
- the possibility to develop allergic reactions.

Few data are present in the literature regarding these two hot points. Further research should be carried out in these directions.

5.2.1. Potential Toxic Compounds in Edible Flowers

To date, there is little information and few studies about the toxicity of edible flowers. The presence of toxic compounds linked to the consumption of flowers can be related to several factors: the type of flower, the cultivation of the flowers, and above all, the presence of toxic compounds in the floral tissues.

Flowers may contain toxic molecules synthetized by the plant to prevent their damage and some of them can act very strongly on humans [117–120]. The quality and quantity of toxic compounds present in plant tissues are very important for the consequences on human health as well as the frequency of consumption. Some inedible flowers contain phytotoxins that can cause serious pathological conditions even if the consumer has eaten small amounts (e.g., *Colchicum autumnale*, *Datura stramonium*, *Nerium oleander*) [121–123]. Most of these plants produce floral nectar consisting of sugars (about 90% by dry weight) and several compounds, including lipids, amino

acids, minerals, antioxidants, and secondary phytotoxic compounds transported between plant tissues via the phloem. Adler [124] summarizes and lists the species that produce nectar containing phytotoxins that are dangerous to bees and humans. The genera that produce nectar harmful to humans are *Agauria*, *Andromeda*, *Kalmia*, *Rododrendon*, *Paullina*, *Azalea*, and *Euphorbia*, and then, the flowers are inedible.

Edible flowers, as well as fruits and vegetables, could contain phytotoxins in small quantities. The ingestion of phytotoxins can give different symptoms such as nausea, vomiting, and diarrhea but also DNA damage that may lead to certain health implications. The recent work of Egebjerg et al. [120] reported a study in the frame of a control campaign by the Danish Veterinary and Food Administration visiting 150 restaurants and local food producers from May to October 2016. The authors studied flowers from 23 plants harvested from the wild, cultivated in private gardens, or in market gardens. The authors [120] presented an evaluation of safety of the selected flowers on the basis of phytochemical studies and toxicological data published in literature: within the 23 flowers, nine reported toxic or potentially toxic compounds, two included unidentified toxic compound(s), and four had potentially toxic compounds found in other plant parts or related species. It is important to underline that also fruits and vegetables could contain a wide variety of plant-derived phytotoxins well listed by Rietjens and collaborators [125] and Pinela et al. [119]; among the various phytotoxins there are aristolochic acid produced by *Aristolochia fanchi*, a herb of folk Chinese medicine; the coumarins present in the essential oils of lavender and mint; pyrrolizine alkaloids contained in the tissues of various plants for food use belonging to the Boraginaceae, Asteraceae, and Fabaceae families; ephedrine alkaloids produced by Ephedra herbs; synephrine, contained in Citrus aurantum and *Citrus reticulata* fruits; kavalactones produced by *Piper methysticum*; anisitin present in *Illicium verum*; cyanogenic glycosides contained in fruits and seeds of several Rosaceae plants (almond, apricot, black cherry, cherry, peach, etc.), sorghum, and cassava; oxalic acid from several wild edible species; saponins from legumes; solanine and chaconine from potatoes; and glycirrhizinic acid present in *Glycyrrhiza glabra* rhizomes.

In edible flowers, the most frequent phytotoxins are thujone and three alkenylbenzenes—methyl eugenol, 1,8 cineole (eucalyptol), and safrole—which are responsible with other aromatic compounds for the taste and aroma of spices and aromatic herbs such as basil, sage, and fennel [125]. These compounds are used to flavor food products, beverages, and cosmetic products or personal hygiene products (e.g., toothpastes, mouthwashes, soaps). Smith and collaborators [126], based on calculations relating to daily exposures to these compounds, believe that they do not pose a significant risk to human health. On the basis of the directives established by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) to identify the tolerable daily dose and the acceptable daily dose of toxic compounds present in food, Egebjerg et al. [120] identify the daily ingestible doses for some edible flowers by adults. The authors indicate for *Achillea millefolium*, an amount of fresh flowers equal to 18 g in order not to exceed the acceptable daily dose of thujone; for *Galium odoratum*, an amount of fresh flowers equal to 7 g in order not to exceed the tolerable daily dose of coumarin; and for *Tropaeolum majus*, an amount of fresh flowers equal to 39.5 g in order not to exceed the tolerable daily dose of erucic acid.

Another possible risk related to the consumption of edible flowers could be represented by the possible concentration of metals and heavy metals in their tissues. In reality, as observed by Grzeszczuk et al. [127] and Drava et al. [128], the risk of metal poisoning, following the consumption of flowers properly grown for human nutrition, was low because the edible flower species were characterized by a relatively low concentration of metals and heavy metals (Cd, Co, Ni, Pb, V). These results were obtained by applying cultivation methods without pesticides and using adequate fertilization. On the other hand, the consumption of edible flowers collected in the field, in polluted areas, and along roadsides, increases the risk of metal poisoning.

It is worth mentioning the review of Lu et al. [129] that showed fifteen years of research (from 2000 to 2015) on common edible flowers; besides traditional use, phytochemicals, and health benefits, the toxicologic aspects were explored. The authors noted that the toxocology of edible flowers is less explored compared to fruits and vegetables and most of the studies analyzed in the

review revealed that edible flowers were non-toxic at an appropriate dosage. However, the authors reported studies on roselle being potentially hazardous [130–133].

5.2.2. Risks Associated With the Consumption of Edible Flowers: Consumer Safety Precautions.

More than one hundred different species produce edible flowers; some of them are commonly consumed unconsciously like artichokes, broccoli, cauliflowers, capers, pumpkin flowers, and saffron pistils; therefore, consumers are not wary of these flower species [129]. Harvesting edible flowers in nature is not recommended without the necessary botanical knowledge and harvesting in urban contexts and on the roadsides is risky for heavy metal contamination. Furthermore, it is not recommended to eat flowers purchased from florists because the ornamental product could be treated with pesticides used to maintain the aesthetic value of cut flowers and pot plants [117]. Safe edible flowers must be cultivated as food, in organic farming or following the rules for vegetable production.

Proper identification of edible flowers is essential to assess safety issues (both toxicological and allergic ones). Moreover, people with asthma or allergies should be cautious because the pollen of specific plants could trigger allergic events. It is important to underline the following issues to take into account:

- the presence of any allergens must be declared to the consumer on the food label or menus, if the meal is eaten away from home;
- to avoid problems related to cross-contamination, especially for allergic people, the hygiene rules in use for all foods must be respected—pollen residues or allergens could give rise to allergic reactions;
- sensitive individuals must gradually introduce new varieties of flowers into their diet to check for any allergic reactions; and
- flowers that are not found to be consumed in Europe before 15 May 1997 must be registered as a novel food.

Few data are present in the literature and further research should be carried out in this direction. For instance, Mazzocchi et al. [134] summarize the main results in the prevention and management of food allergies, highlighting the key role of nutritional components and dietary habits in the inflammatory response of the immune system. The same authors marked how individualized strategies should be implemented in terms of food allergy management; these strategies will include developmental preparation for weaning, the prevalence of particular food allergies in some countries, family eating habits, and the availability of medical and dietary care. Costa et al. [135], regarding the management of food allergies, with particular attention on natural tolerance and food sensitization acquisition, underline how immunotherapy treatments can be an innovative approach in this field. Although the topic is still debated, these treatments have been found to be effective, safe, and adjuvant in patients with multiple food allergies.

5.3. A Case Study: The ANTEA Project

In this scenario, in the framework of the project named "Innovative activities for the development of the cross-border supply chain of the edible flower" (ANTEA), the potential allergenicity of examples of edible flowers was studied.

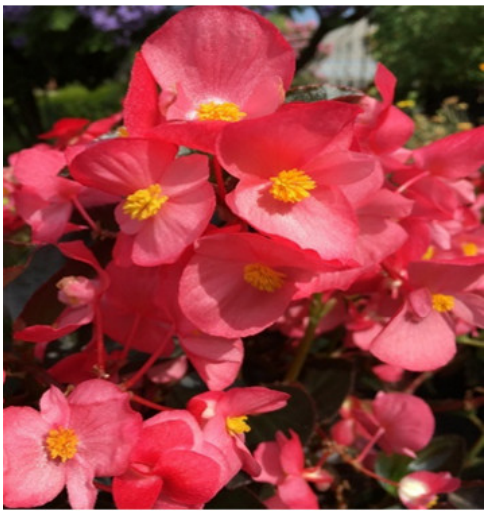
The project ANTEA is concentrated on the development and the implementation of the edible flower chain, from production with sustainable methods to distribution, conservation, safe use, and the study of chemical and organoleptic characteristics of a sample of 40 species.

In Supplementary file S1, 62 edible flowers are described, selected for the study of allergenicity, family, genus, species, variety, origin, and English and Italian names. Moreover, Supplementary file S1 well summarized their application in nutrition and phytochemistry evidence and documented references.

The species and varieties covered by the project were selected on the basis of various factors such as stable presence on the market as edible flowers, use in traditional recipes in Europe or in the rest of the world, organoleptic and/or decorative characteristics, and information already present in

the literature. Some species such as violets and pumpkin flowers are already marketed in the supermarket in polyethylene bags or mixed with salads [103]. Many flowers used to produce syrups, liqueurs, and herbal teas can also be used fresh. This is the case of the different varieties of roses, hibisci, calendulas, and cornflowers. Even some flowers used for the preparation of perfumes are edible; they contain essential oils in petals that confer an intense taste to rose, lavender, tuberose, and carnation flowers. Wild flowers such as dandelions, borages, daisies, primroses, snapdragons, nasturtia (Figure 4A), wild garlic, and some species of clover are reported in ancient and traditional regional recipes mainly in soups and fresh salads. Some of these flowers have satisfying colors, forms, and interesting tastes which is the reason why many chefs and confectioners use edible flowers to add unusual flavors and visual appeal to their creations. Cooking with edible flowers is reported in many extra-European nations. For example, the inflorescences of *Acmella oleracea* are spicy, sparkling, and "anesthetize" the mouth as piment; they are used in South America raw or cooked for traditional salads and stews [136]. In South Africa, *Tulbaghia* flowers are called "social garlic" because they have a pleasant garlic taste and are perfectly digestible. In Eastern countries, flowers such as *chrysanthemum* and butterfly pea are commonly used. In regards to taste, begonias (Figure 4B) give a light acidic note to dishes, as well as *Pelargonium* odoratum "Lemon" enriches food with a pleasant lemon aroma; flowers and leaves of *Mertensia maritima* give a special oyster flavor [137]. Other flowers that normally decorate terraces and balconies, properly grown, can have another utilization; it is the case of floss flowers, marigolds, petunias, and fuchsias with carrot, spicy, floral, and radish tastes, respectively. Attractive flowers involved in garden design such as *Verbena bonariensis*, dahlias, faassen's catmint, and daylilies can be eaten in salads. Finally, the flowers of many aromatic and spice species can be eaten, giving the characteristic fragrance present in the glandular trichomes; the sage, basil, and rosemary flowers have the same taste as the leaves but without the bitter notes. Some species and varieties of *Agastache* are very sweet and aromatic (licorice, mint, lemon, and anise) and rich in secondary compounds [138]. Monarda dark red petals (Figure 4C) can be used fresh or dried to garnish pizza or pasta due to their oregano and bergamot taste. Andean sage (*Salvia discolor*—Figure 4D) produces flowers with abundant nectar and petals with black currant and pine nut tastes. These edible flowers are rich in secondary compounds and antioxidants [139] that can enrich the human diet. In addition, the tepals of *Crocus sativus*, bio-residues of the saffron economy, are completely edible and can be used fresh or dried providing plant secondary compounds (polyphenols, anthocyanins, and flavonoids) with good antioxidant activity [140].

(a)



(b)

(c)

(d)

Figure 4. Four examples of edible flowers investigated during the "Innovative activities for the development of the cross-border supply chain of the edible flower" (ANTEA) project: (**a**) *Tropaeolum majus*, (**b**) Begonia semperflorens, (**c**) *Monarda didima*, (**d**) *Salvia discolor*.

In the ANTEA project, cytotoxicity tests were performed on two immortalized cell lines commonly used for this biomedical test: COS-7 from monkey kidney and HepG2 from the human liver. The two cell lines were cultured for 24 h in the presence of the edible flower extracts at five different concentrations (1000, 100, 10, 1, and 0.1 μg/mL). The cells exposed to the flower extracts of eight species or varieties (*Acmella oleracea*, *Ageratum houstonianum*, Begonia semperflorens, *Fuchsia regia*, *Ocimum basilicum* "citriodorum", *Tulbaghia cominsii*, *Tulbaghia violacea*, T. violacea "Alba") showed a cell vitality percentage of less than 50% at a concentration of 1000 μg/mL [141].

Concerning the potential allergenicity of edible flowers studied in the ANTEA project, we worked in two consecutive steps: bibliographic research and allergen-dedicated database consultation.

Bibliographic research about edible flowers has highlighted that few data are present in the literature. No data have been found in the literature that reports the presence of allergens belonging to the superfamily of the plant kingdom.

Database Allergen Nomenclature and the COMprehensive Protein Allergen REsource (COMPARE) Database were used to identify possible allergens in the selected flower sample of the ANTEA project. The searching was carried out by entering the scientific name of the potential allergen source and covering the different anatomical plant parts. The selected flower sample and related allergens identified are reported as Supplementary file S2.

By searching in the COMPARE Database for *Crocus sativus* referred to saffron pollen and stamen [142], we have found the allergen profilin protein. The Allergen Nomenclature Database reported for *Crocus sativus* two allergen proteins: Cro s 1 and Cro s 2 (profiling) respectively with molecular weights of 21 and 14 kDa.

By using Allergen Nomenclature for another species of genus *Cucurbita*, *Curcurbita maxima*, we found a positive match with two allergens: Cuc ma 4 (11S globulin, 50 kDa) and Cuc ma 5 (2S albumin, 14 kDa). This result gives us an alert for Cucurbita sp. pepo.

The results obtained by consulting the two databases considered, COMPRISE and Allergen Nomenclature, marked two alerts for two varieties of edible flowers (see Supplementary file S2) selected in the project. It is necessary to consider not only the direct allergic reactions, but also the cross-reactions that can occur (see especially the *Crocus sativus* case).

However, it is important to underline that the databases considered do not allow us to conclude that the selected varieties do not contain allergens, but only that at present there is no scientific evidence that proves otherwise.

Another safety parameter is a clear and verified documentation on the historical consumption of the edible flower. Ongoing activity is verifying if the selected flowers sample of the ANTEA project may be considered novel foods on the basis of a significant consumption in Europe prior to 15 May 1997 (Regulation (EC) 258/97 on novel food and novel food ingredients). The edible flowers of the ANTEA project which present consumption data prior to May 1997 (and, therefore, not considered as novel food) can be considered safe from the allergenic point of view. The other edible flowers of which it is not possible to demonstrate a historical consumption prior to May 1997 must be submitted to the EFSA for recognition as novel food.

However, it should be considered, as part of the safety assessment, that edible flowers are generally consumed in composite dishes in quantities of the order of grams. It should also be considered that if the product is processed (i.e., cooking procedures), the allergenic potential could have significant variations.

As for labeling, the edible flowers selected for the ANTEA project not belonging to the 14 main allergenic food groups do not need to be highlighted in the product label. In the case of cross-contamination, the edible flowers follow the regulation of other foods (see Figure 3).

6. Conclusions

Edible flowers are and have been used in some cultures for a long time; nonetheless, nowadays they have become a novel trend in food. The demand for edible flowers expanded worldwide in recent years, answering to requests of both consumers and chefs which suggests that edible flowers are safe, may improve flavor, give color, and enhance the visual appearance of dishes. The main current challenge is to define a shared regulation system to assess the context, safety, and use of edible flowers. Considering allergy aspects it is important to underline that new edible flower must be introduced in the EU market following the European regulation for novel foods [93].

Supplementary Materials: The following are available online at www.mdpi.com/2071-1050/12/20/8709/s1, Supplementary file S1: Description and phytochemistry evidence of 62 edible flowers; Supplementary file S2: potentially allergenicity evaluation of 62 edible flowers.

Author Contributions: Conceptualization, M.L., E.L. and B.R.; Data curation, A.D., M.L., A.C., P.G.; Writing of the original manuscript, M.L., A.C., A.D., G.L.-B., E.L., A.S., B.R.; Review and editing of the manuscript, M.L., A.C., A.D., P.G., G.L.-B., E.L., A.S., B.R.; Project Administration, B.R.; All authors have contributed to, read, and agreed to the published version of the manuscript.

Funding: This research was funded by the ANTEA Project (Attività innovative per lo sviluppo della filiera transfrontaliera del fiore edule (n. 1139)), grant number CUP C12F17000080003 (INTERREG-ALCOTRA UE 2014–2020).

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Andreas, L. Allergenicity of food and impact of processing. In *Novel Food Processing*; CRC Press: Boca Raton, FL, USA, 2009; pp. 459–478**.**
2. Sicherer, S.H.; Sampson, H.A. Peanut allergy: Emerging concepts and approaches for an apparent epidemic. *J. All. Clin. Immunol.* **2007**, *120*, 491–503.
3. Valenta,R.; Hochwallner, H.; Linhart, B.; Pahr, S. Food Allergies: The Basics. *Gastroenterology* **2015**, *148*, 1120–1131, doi:10.1053/j.gastro.2015.02.006.
4. Sathe, S.K.; Liu, C.; Zaffran, V.D. Food Allergy. *Annu. Rev. Food Sci. Technol.* **2016**, *7*, 191–220, doi:10.1146/annurev-food-041715-033308.
5. Nuray Bayar Muluk, M.D.; Cemal Cingi, M.D. Oral allergy syndrome. *Am. J. Rhinol. Allergy* **2018**, *32*, 27–30, doi:10.2500/ajra.2018.32.4489.
6. Iweala, O.I.; Choudhary, S.K.; Commins, S.P. Food Allergy. *Curr. Gastroenterol. Rep.* **2018**, *20*, 17, doi:10.1007/s11894-018-0624.
7. Jimenez-Rodriguez, T.W.; Garcia-Neuer, M.; Alenazy, L.A.; Castells, M. Anaphylaxis in the 21st century: Phenotypes, endotypes, and biomarkers. *J. Asthma Allergy* **2018**, 11, 121–142, doi:10.2147/JAA.S159411.

8. Yu, W.; Freeland, D.M.H.; Nadeau, K.C. Food allergy: Immune mechanisms, diagnosis and immunotherapy. *Nat. Rev. Immunol.* **2016**, *16*, 751–765, doi:10.1038/nri.2016.111.
9. Waserman, S.; Bégin, P.; Watson, W. IgE-mediated food allergy. *Allergy Asthma Clin. Immunol.* **2018**, *14*, 55, doi:10.1186/s13223-018-0284-3.
10. Anvari, S.; Miller, J.; Yeh, C.Y.; Davis, C.M. *Clinic. Rev. Allergy Immunol.* **2019**, *57*, 244, doi:10.1007/s12016-018-8710-3.
11. Nowak-Węgrzyn, A.; Katz, Y.; Mehr, S.S.; Koletzko, S. Non-IgE-mediated gastrointestinal food allergy. *Allergy Asthma Clin. Immunol.* **2018**, *14*, 55, doi: 10.1016/j.jaci.2015.03.025.
12. Burton, O.T.; Oettgen, H.C. Beyond immediate hypersensitivity: Evolving roles for IgE antibodies in immune homeostasis and allergic diseases. *Immunol Rev.* **2011**, *242*, 128–143, doi:10.1111/j.1600-065X.2011.01024.x.
13. Galli, S.J.; Tsai, M. IgE and mast cells in allergic disease. *Nat. Med*. **2012**, *18*, 693–704, doi:10.1038/nm.2755.
14. Sicherer, S.H.; Sampson, H.A. Food allergy: Epidemiology, pathogenesis, diagnosis, and treatment. *J. Allergy Clin. Immunol.* **2014**, *133*, 291–307, quiz 8. Epub 2014/01/07, doi:10.1016/j.jaci.2013.11.020.
15. Hogan, S.P.; Wang, Y.H.; Strait, R.; Finkelman, F.D. Food-induced anaphylaxis: Mast cells as modulators of anaphylactic severity. *Semin. Immunopathol.* **2012**, *34*, 643–653, doi:10.1007/s00281-012-0320-1.
16. Groschwitz, K.R.; Hogan, S.P. Intestinal barrier function: Molecular regulation and disease pathogenesis. *J. Allergy Clin. Immunol.* **2009**, *124*, 3–22.
17. Ferreira, F.; Hawranek, T.; Gruber, P.; Wopfner, N.; Mari, A. Allergic cross-reactivity: From gene to the clinic. *Allergy* **2004**, *59*, 243–267, doi:10.1046/j.1398-9995.2003.00407.x.
18. Canonica, G.W.; Ansotegui, I.J.; Pawankar, R.; Schmid-Grendelmeier, P.; van Hage, M.; Baena-Cagnani, C.E.; Melioli, G.; Nunes, C.; Passalacqua, G.; Rosenwasser, L.; et al. A WAO—ARIA—GALEN consensus document on molecular-based allergy diagnostics. *World Allergy Organ J.* **2013**, *6*,17.
19. Sampson, H.A.; Aceves, S.; Bock, S.A.; James, J.; Jones, S.; Lang, D.; Nadeau, K.; Nowak-Wegrzyn, A.; Oppenheimer, J.; Perry, T.T.; et al. Food allergy: A practice parameter update-2014. *J. Allergy Clin. Immunol.* **2014**, *134*, 1016–1025.
20. Popescu, F.D. Cross-reactivity between aeroallergens and food allergens. *World J. Methodol.* **2015**, *5*, 31–50, doi:10.5662/wjm.v5.i2.31.
21. Egger, M.; Mutschlechner, S.; Wopfner, N.; Gadermaier, G.; Briza, P.; Ferreira, F. Pollen-food syndromes associated with weed pollinosis: An update from the molecular point of view. *Allergy* **2006**, *61*, 461–476.
22. Han, Y.; Kim, J.; Ahn, K. Food allergy. *Korean J. Pediatr.* **2012**, *55*, 153–158.
23. Popescu, F.D. Molecular biomarkers for grass pollen immunotherapy. *World J. Methodol.* **2014**, *4*, 26–45.
24. Werfel, T.; Asero, R.; Ballmer-Weber, B.K.; Beyer, K.; Enrique, E.; Knulst, A.C.; Mari, A.; Muraro, A.; Ollert, M.; Poulsen, L.K.; et al. Position paper of the EAACI: Food allergy due to immunological cross-reactions with common inhalant allergens. *Allergy* **2015**, *70*, 1079–1090, doi:10.1111/all.12666.
25. Sharma, H.P.; Bansil, S.; Uygungil, B. Signs and symptoms of food allergy and food-induced anaphylaxis. *Ped. Clin. N. Am.* **2015**, *62*, 1377–1392.
26. Oriel, R.C.; Wang, J.J. Diagnosis and management of food allergy. *Ped. Clin. N. Am.* **2019**, *66*, 941–954.
27. Hoffmann-Sommergrube, K.; Mills, E.N. Food allergen protein families and their structural characteristics and application in component-resolved diagnosis: New data from the EuroPrevall project. *Anal. Bioanal. Chem.* **2009**, *395*, 25–35, doi:10.1007/s00216-009-2953-z.
28. Radauer, C.; Bublin, M.; Wagner, S.; Mari, A.; Breiteneder, H. Allergens are distributed into few protein families and possess a restricted number of biochemical functions *J. Allergy Clin. Immunol.* **2008**, *121*, 847–852.
29. Jenkins, J.A.; Griffiths-Jones, S.; Shewry, P.R.; Breiteneder, H.; Mills, E.N. Structural relatedness of plant food allergens with specific reference to cross-reactive allergens: An in silico analysis. *J. Allergy Clin. Immunol.* **2005**, *115*, 163–170.
30. Jenkins, J.A.; Breiteneder, H.; Mills, E.N. Evolutionary distance from human homologs reflects allergenicity of animal food proteins. *J. Allergy Clin. Immunol.* **2007**, *120*, 1399–1405.
31. Hoffmann-SommergruberRadauer, C.; Bublin, M.; Wagner, S.; Mari, A.; Breiteneder, H. Allergens are distributed into few protein families and possess a restricted number of biochemical functions. *J. Allergy Clin. Immunol*. **2008**, 121, 847-852.
32. Breiteneder, H.; Mills, E.N. Molecular properties of food allergens. *J. Allergy Clin. Immunol.* **2005**, *115*, 14–23.

33. Dall'Antonia, F.; Pavkov-Keller, T.; Zangger, K.; Keller, W. Structure of allergens and structure based epitope predictions. *Methods* **2014**, *66*, 3–21, doi:10.1016/j.ymeth.2013.07.024.
34. FDA. *The Threshold Working Group. Report. Approaches to Establish Thresholds for Major Food Allergens and for Gluten in Food*; FDA: Silver Spring, MD, USA, 2006. Available online: https://www.fda.gov/media/78205/download.
35. Blom, W.M.; Vlieg-Boerstra, B.J.; Kruizinga, A.G.; van der Heide, S.; Houben, G.F.; Dubois, A.E. Threshold dose distributions for 5 major allergenic foods in children. *J. Allergy Clin. Immunol.* **2013**, *131*, 172–179, doi:10.1016/j.jaci.2012.10.034.
36. Council of the European Union. Directive 2003/89/EC (and further amendments) introduced Annex IIIa. *Off. J. Eur. Union* **2003.** Available at: https://www.fsai.ie/uploadedFiles/Dir2003.89.pdf (accessed on 18 September 2020).
37. Allergen Nomenclature. Available online: http://www.allergen.org/ (accessed on 12 July 2020).
38. COMprehensive Protein Allergen Resource. COMPARE 2020 DB Release Date: 01/29/2020. Available online: https://comparedatabase.org/ (accessed on 12 July 2020).
39. Tong, J.C.; Lim, S.J.; Muh, H.C.; Chew, F.T.; Tammi, M.T. Allergen Atlas: A comprehensive knowledge center and analysis resource for allergen information. *Bioinformatics* **2009**, *25*, 979–980, doi:10.1093/bioinformatics/btp077.
40. Radauer, C. Navigating through the Jungle of Allergens: Features and Applications of Allergen Databases. *Int. Arch. Allergy Immunol.* **2017**, *173*, 1–11, doi:10.1159/000471806.
41. Allergome. Available online: http://www.allergome.org/script/about.php (accessed on 12 July 2020).
42. AllergenOnline. Available online: http://www.allergenonline.org/ (accessed on 12 July 2020).
43. Structural Database of Allergenic Proteins (SDAP). Available online: https://fermi.utmb.edu/ (accessed on 12 July 2020).
44. Immune Epitope Database (IEDB). Available online: https://www.iedb.org/ (accessed on 12 July 2020).
45. Lorenz, A.R.; Scheurer, S.; Vieths, S. Food allergens: Molecular and immunological aspects, allergen databases and cross-reactivity. *Chem. Immunol. Allergy* **2015**, *101*, 18–29, doi:10.1159/000371647.
46. Pekar, J.; Ret, D.; Untersmayr, E. Stability of allergens. *Mol. Immunol.* **2018**, *100*, 14–20, doi:10.1016/j.molimm.2018.03.017.
47. Amarowicz, R.; Carle, R.; Dongowski, G.; Durazzo, A.; Galensa, R.; Kammerer, D.; Maiani, G.; Piskula, M.K. Influence of postharvest processing and storage on the content of phenolic acids and flavonoids in foods. *Mol. Nutr. Food Res.* **2009**, *53*, S151–S183.
48. Nicoli, M.C.; Anese, M.; Parpinel, M. Influence of processing on the antioxidant properties of fruit and vegetables. *Trends Food Sci. Technol.* **1999**, *10*, 94–100.
49. Thomas, K.; Herouet-Guicheney, C.; Ladics, G.; Bannon, G.; Cockburn, A.; Crevel, R.; Fitzpatrick, J.; Mills, C.; Privalle, L.; Vieths, S. Evaluating the effect of food processing on the potential human allergenicity of novel proteins: International workshop report. *Food Chem. Toxicol.* **2007**, *45*, 1116–1122.
50. Mills, E.N.C.; Mackie, A.R. The impact of processing on allergenicity of food. *Curr. Opin. Allergy Clin. Immunol.* **2008**, *8*, 249–253.
51. Jimenez-Saiz, R., S.; Benede, E.; Molina, Lopez-Exposito. I. Effect of processing technologies on the allergenicity of food products. *Crit. Rev. Food Sci. Nutr.* **2015**, *55*, 1902–1917.
52. Lepski, S.; Brockmeyer, J. Impact of dietary factors and food processing on food allergy. *Mol. Nutr. Food Res.* **2013**, *57*, 145–152, doi:10.1002/mnfr.201200472.
53. EFSA, 2014. Scientific opinion on the evaluation of allergenic foods and food ingredients for labeling purposes. *EFSA J.* **2014**, *12*, 3894.
54. Verhoeckx, K.C.M.; Vissers, Y.M.; Baumert, J.L.; Faludi, R.; Feys, M.; Flanagan, S.; Herouet-Guicheney, C.; Holzhauser, T.; Shimojo, R.; van der Bolt, N.; et al. Food processing and allergenicity. *Food Chem. Toxicol.* **2015**, *80*, 223–240.
55. Vanga, S.K.; Singh, A.; Raghavan, V. Review of conventional and novel food processing methods on food allergens. *Crit. Rev. Food Sci. Nutr.* **2017**, *57*, 2077–2094, doi:10.1080/10408398.2015.1045965.
56. Cabanillas, B.; Novak, N. Effects of daily food processing on allergenicity. *Crit. Rev. Food Sci. Nutr.* **2017**, *59*, 31–42, doi:10.1080/10408398.2017.1356264.
57. Mills, E.N.C.; Sancho, A.; Kostyra, H. The effect of food processing on allergens. In *Managing Allergens in Food*; Hoffmann-Somergruber, K., Mills, C., Wichers, H.J., Eds.; Woodhead Publishing: Cambridge, UK, 2006.

58. Cabanillas, B.; Jappe, U.; Novak, N. Allergy to peanut, soybean, and other legumes: Recent advances in allergen characterization, stability to processing and IgE cross-reactivity. *Mol. Nutr. Food Res.* **2018**, *62*, doi:10.1002/mnfr.201700446.
59. Sathe, S.K.; Teuber, S.S.; Roux, K.H. Effects of food processing on the stability of food allergens. *Biotechnol. Adv.* **2005**, *23*, 423–429.
60. Andreas, L Sathe S.K,; Sharma G.M. Effects of food processing on food allergens. *Mol Nutr Food Res*. **2009**, 53, 970-978.
61. Rahaman, T.; Vasiljevic, T.; Ramchandran, L. Effect of processing on conformational changes of food proteins related to allergenicity. *Trends Food Sci. Technol.* **2016**, *49*, 24–34, doi:10.1016/j.tifs.2016.01.001.
62. Ahrazem, O.; Jimeno, L.; Lopez-Torrejon, G.; Herrero, M.; Espada, J.L.; Sanchez-Monge, R.; Duffort, O.; Barber, D.; Salcedo, G. Assessing allergen levels in peach and nectarine cultivars. *Ann. All. Asthma Immunol.* **2007**, *99*, 42–47.
63. Boyano-Martınez, T.; Pedrosa, M.; Belver, T.; Quirce, S.; Garcıa-Ara, C. Peach allergy in Spanish children: Tolerance to the pulp and molecular sensitization profile. *Pediat. All. Immunol.* **2013**, *24*, 168–172.
64. Pravettoni, V.; Primavesi, L.; Farioli, L.; Brenna, O.V.; Pompei, C.; Conti, A.; Scibilia, J.; Piantanida, M.; Mascheri, A.; Pastorello, E.A. Tomato allergy: Detection of IgE-binding lipid transfer proteins in tomato derivatives and in fresh tomato peel, pulp, and seeds. *J. Agric. Food Chem*. **2009**, *57*, 10749–10754.
65. Wang, J.; Vanga, S.K.; Raghavan, V. Effect of pre-harvest and post-harvest conditions on the fruit allergenicity: A review. *Crit. Rev. Food Sci. Nutr.* **2019**, *59*, 1027–1043, doi:10.1080/10408398.2017.1389691.
66. Tulipani, S.; Marzban, G.; Herndl, A.; Laimer, M.; Mezzetti, B.; Battino, M. Influence of environmental and genetic factors on healthrelated compounds in strawberry. *Food Chem.* **2011**, *124*, 906–913.
67. Lopez-Matas, M.A.; Larramendi, C.H.; Ferrer, A.; Huertas, A.J.; Pagan, J.A.; Garcıa-Abujeta, J.L.; Dolle, S.; Lehmann, K.; Schwarz, D.; Weckwert, W.; et al. Allergenic activity of different tomato cultivars in tomato allergic subjects. *Clin. Exp. Allergy* **2011**, *41*, 1643–165.
68. Schmitz-Eiberger, M.; Matthes, A. Effect of harvest maturity, duration of storage and shelf life of apples on the allergen Mal d 1, polyphenoloxidase activity and polyphenol content. *Food Chem.* **2011**, *127*, 1459–1464.
69. Wang, J.; Liang, S.; Ma, H.; Zhang, P.; Shi, W. Effects of ethephon on fresh In-Husk walnut preservation and its possible relationship with phenol metabolism. *J. Food. Sci.* **2016**, *81*, C1921–C1927.
70. Teodorowicz, M.; van Neerven, J.; Savelkoul, H. Food processing: The influence of the maillard reaction on immunogenicity and allergenicity of food proteins. *Nutrients* **2017**, *9*, 835, doi:10.3390/nu9080835www.mdpi.com/journal/nutrients.
71. Gupta, R.K.; Gupta, K.; Sharma, A.; Das, M.; Ansari, I.A.; Dwivedi, P.D. Maillard reaction in food allergy: Pros and cons. *Crit. Rev. Food Sci. Nutr.* **2018**, *58*, 208–226, doi:10.1080/10408398.2016.1152949.
72. Toda, M.; Hellwig, M.; Henle, T.; Vieths, S. Influence of the maillard reaction on the allergenicity of food proteins and the development of allergic inflammation. *Curr. Allergy Asthma Rep.* **2019**, *19*, 4, doi:10.1007/s11882-019-0834-x.
73. Heilmann, M.; Wellner, A.; Gadermaier, G.; Ilchmann, A.; Briza, P.; Krause, M.; Henle, T. Ovalbumin modified with pyrraline, a Maillard reaction product, shows enhanced T-cell immunogenicity. *J. Biol. Chem.* **2014**, *289*, 7919–7928.
74. Gruber, P.; Becker, W.M.; Hofmann, T. Influence of the Maillard reaction on the allergenicity of rAra h 2, a recombinant major allergen from peanut (*Arachis hypogaea*), its major epitopes, and peanut agglutinin. *J. Agric. Food Chem.* **2005**, *53*, 2289–2296.
75. Seo, S.; L'Hocine, L.; Karboune, S. Allergenicity of potato proteins and of their conjugates with galactose, galactooligosaccharides, and galactan in native, heated, and digested forms. *J. Agric. Food Chem.* **2014**, *62*, 3591–3598.
76. De Jongh, H.H.; Robles, C.L.; Timmerman, E.; Nordlee, J.A.; Lee, P.W.; Baumert, J.L.; Hamilton, R.G.; Taylor, S.L.; Koppelman, S.J. Digestibility and IgE-binding of glycosylated codfish parvalbumin. *Biomed. Res. Int.* **2013**, *2013*, 756789.
77. Rupa, P.; Nakamura, S.; Katayama, S.; Min, Y. Effects of ovalbumin glycoconjugates on alleviation of orally induced egg allergy in mice via dendritic-cell maturation and T-cell activation. *Mol. Nutr. Food Res.* **2014**, *58*, 405–417.
78. Rupa, P.; Nakamura, S.; Katayama, S.; Mine, Y. Attenuation of allergic immune response phenotype by mannosylated egg white in orally induced allergy in BALB/c mice. *J. Agric. Food Chem*. **2014**, *62*, 9479–9487.

79. Kroghsbo, S.; Rigby, N.M.; Johnson, P.E.; Adel-Patient, K.; Bøgh, K.L.; Salt, L.J.; Mills, E.C. and Madsen, C.B. Assessment of the sensitizing potential of processed peanut proteins in Brown Norway rats: Roasting does not enhance allergenicity. *PLoS ONE* **2014**, *7*, e96475, doi:10.131471/journal.pone.0096475.
80. Iwan, M.; Vissers, Y.M.; Fiedorowicz, E.; Kostyra, H.; Kostyra, E.; Savelkoul, H.F.; Wichers, H.J. Impact of Maillard reaction on immunoreactivity and allergenicity of the hazelnut allergen Cor a 11. *J. Agric. Food Chem.* **2011**, *59*, 7163–7171.
81. Verma, A.K.; Kumar, S.; Das, M.; Dwivedi, P.D. Impact of thermal processing on legume allergens. *Plant Foods Hum. Nutr.* **2012**, *67*, 430–441, doi:10.1007/s11130-012-0328-7.
82. Cuadrado, C.; Cabanillas, B.; Pedrosa, M.M.; Varela, A.; Guillamon, E.; Muzquiz, M.; Crespo, J.F.; Rodriguez, J.; Burbano, C. Influence of thermal processing on IgE reactivity to lentil and chickpea proteins. *Mol. Nutr. Food Res.* **2009**, *53*, 1462–1468.
83. Pastorello, E.A.; Pravettoni, V.; Farioli, L.; Primavesi, L.; Scibilia, J.; Piantanida, M.; Mascheri, A.; Conti, A. Green bean (*Phaseolus vulgaris*): A new source of IgE-binding lipid transfer protein. *J. Agric. Food Chem.* **2009**, *58*, 4513–4516.
84. Kasera, R.; Singh, A.B.; Lavasa, S.; Nagendra, K.; Arora, N. Purification and immunobiochemical characterization of a 31 kDa crossreactive allergen from *Phaseolus vulgaris* (kidney bean). *PLoS ONE* **2013**, *8*, e63063.
85. Vitaliti, G.; Morselli, I.; Di Stefano, V.; Lanzafame, A.; La Rosa, M.; Leonardi., S. Urticaria and anaphylaxis in a child after inhalation of lentil vapours: A case report and literature review. *Ital. J. Pediatr.* **2012**, *38*, 71.
86. Chang, C.; Lahti, T.; Tanaka, T.; Nickerson, M.T. Egg proteins: Fractionation, bioactive peptides and allergenicity. *J. Sci. Food Agric.* **2018**, *98*, 5547–5558, doi:10.1002/jsfa.9150.
87. Esteghlal, S.; Gahruie, H.H.; Niakousari, M.; Barba, F.J.; Bekhit, A.E.; Mallikarjunan K5, Roohinejad, S. Bridging the knowledge gap for the impact of non-thermal processing on proteins and amino acids. *Foods* **2019**, *8*, E262, doi:10.3390/foods8070262.
88. Huang, H.W.; Hsu, C.P.; Yang, B.B.; Wang, C.Y. Potential utility of high-pressure processing to address the risk of food allergen concerns. *Compr. Rev. Food Sci. Technol.* **2014**, *13*, 78–90.
89. Yang, A.; Zuo, L.; Cheng, Y.; Wu, Z.; Li, X.; Tong, P.; Chen, H. Degradation of major allergens and allergenicity reduction of soybean meal through solid-state fermentation with microorganisms. *Food Funct.* **2018**, *9*, 1899–1909, doi:10.1039/c7fo01824j. .
90. Council of the European Union. Directive 2000/13/EC of the European Parliament and of the Council of 20 March 2000 on the approximation of the laws of the Member States relating to the labelling, presentation and advertising of foodstuffs. *Off. J. Eur. Union* **2000.** Available at: https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32000L0013&from=EN (accessed on 18 September 2020).
91. Council of the European Union. COMMISSION DIRECTIVE 2007/68/EC of 27 November 2007 amending Annex IIIa to Directive 2000/13/EC of the European Parliament and of the Council as regards certain food ingredients. *Off. J. Eur. Union* **2007**. Available at: http://extwprlegs1.fao.org/docs/pdf/eur75647.pdf (accessed on 18 September 2020).
92. Council of the European Union. Regulation (EU) No 1169/2011 of the European Parliament and of the Council of 25 October 2011 on the provision of food information to consumers. *Off. J. Eur. Union* **2011**. Available at: https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32011R1169&from=EN (accessed on 18 September 2020).
93. Council of the European Union. Regulation EC No. 258/97 of the European Parliament and of the Council of 27 January 1997 concerning novel foods and novel food ingredients. *Off. J. Eur. Comm.* **1997**, *40*, L43. ISSN 0378-6978.
94. European Commission. Regulation (EU) 2015/2283 of the European Parliament and of the Council of 25 November 2015 on novel foods. Available online: https://ec.europa.eu/food/safety/novel_food/legislation_en.
95. European Commission. List containing all authorised novel foods (Commission Implementing Regulation (EU) 2017/2470). Available online: https://ec.europa.eu/food/safety/novel_food/authorisations/union-list-novel-foods_en (accessed on 20 September 2020).

96. European Commission. Novel Food Catalogues. Available online: https://ec.europa.eu/food/safety/novel_food/catalogue/search/public/index.cfm (accessed on 20 September 2020).
97. Cunningham, E. What nutritional contribution do edible flowers make? *J. Acad. Nutr. Diet* **2015**, *115*, 856, doi:10.1016/j.jand.2015.03.002.
98. Murphy, H. Foods indigenous to the Western hemisphere. In *American Indian Health Diet Project*; Available online: http://www.aihd.ku.edu/ foods/squash.html (accessed on 18 September 2020).
99. Kirker, C.L.; Newman, M. *Edible Flowers: A Global History*, 1st ed.; Reaktion Books Ltd.; London, UK, 2016; pp. 19–21.
100. Stradley, L. Edible flowers are the new rage in haute cuisine. In *What's Cooking America Website*; Available online: http://whatscookingamerica.net/EdibleFlowers/EdibleFlowersMain.htm (accessed on 27 February 2015).
101. Falconnier, D. *Incredible Edible Flowers*; University of Illinois Extension: Urbana, IL, USA, 2006; pp 1–6.
102. Mlcek, J.; Rop, O. Fresh edible flowers of ornamental plants—A new source of nutraceutical foods. *Trends Food Sci. Technol.* **2011**, *22*, 561–569, doi:10.1016/j.tifs.2011.04.006.
103. Fernandes, L.; Casal, S.; Pereira, J.A.; Saraiva, J.A.; Ramalhosa, E. An Overview on the market of edible flowers. *Food Rev. Int.* **2019**, *36*, 258–275; doi:10.1080/87559129.2019.1639727.
104. Rop, O.; Mlcek, J.; Jurikova, T.; Neugebauerova, J.; Vabkova, J. Edible flowers—A new promising source of mineral elements in human nutrition. *Molecules* **2012**, *17*, 6672–6683.
105. Chensom, S.; Okumura, H.; Mishima, T. Primary screening of antioxidant activity, total polyphenol content, carotenoid content, and nutritional composition of 13 edible flowers from Japan. *Prev. Nutr. Food Sci.* **2019**, *24*, 171–178, doi:10.3746/pnf.2019.24.2.171.
106. Lee, J.H.; Lee, H.J.; Choung, M.G. Anthocyanin compositions and biological activities from the red petals of Korean edible rose (*Rosa hybrida* cv. *Noblered*). *Food Chem.* **2011**, *129*, 272–278.
107. Zhang, L.; Yang, X.; Zhang, Y.; Wang, L.; Zhang, R. In vitro antioxidant properties of different parts of pomegranate flowers. *Food Bioprod. Process.* **2011**, *89*, 234–240, doi:10.1016/j.fbp.2010.04.007.
108. Arya, V.; Kumar, D.; Gautam, M. Phytopharmacological review on flowers: Source of inspiration for drug discovery. *Biomed. Prev. Nutr.* **2014**, *4*, 45–51, doi:10.1016/j.bionut.2013.08.009.
109. Zeng, Y.; Deng, M.; Lv, Z.; Peng, Y. Evaluation of antioxidant activities of extracts from 19 Chinese edible flowers. *SpringerPlus* **2014**, *3*, 315, doi:10.1186/2193-1801-3-315.
110. Pires, T.C.; Dias, M.I.; Barros, L.; Ferreira, I.C. Nutritional and chemical characterization of edible petals and corresponding infusions: Valorization as new food ingredients. *Food Chem.* **2017**, *220*, 337–343, doi:10.1016/j.foodchem.2016.10.026.
111. Pires, T.C.S.P.; Dias, M.I.; Barros, L.; Calhelha, R.C.; Alves, M.J.; Oliveira, M.B.P.P.; Santos-Buelga, C.; Ferreira, I.C.F.R. Edible flowers as sources of phenolic compounds with bioactive potential. *Food Res. Int.* **2018**, *105*, 580–588, doi:10.1016/j.foodres.2017.11.014. .
112. Nowicka, P.; Wojdyło, A. Anti-hyperglycemic and anticholinergic effects of natural antioxidant contents in edible flowers. *Antioxidants* **2019**, 8, E308, doi:10.3390/antiox8080308.
113. Zhao, L.; Fan, H.; Zhang, M.; Chitrakar, B.; Bhandari, B.; Wang, B. Edible flowers: Review of flower processing and extraction of bioactive compounds by novel technologies. *Food Res. Int.* **2019**, *126*, 108660, doi:10.1016/j.foodres.2019.108660.
114. US Department of Agriculture, Agricultural Research Service. USDA National Nutrient Database for Standard Reference, Release 27. Available online: http://www.ars.usda.gov/Services/docs..htm?docid¼8964 (accessed on 8 March 2015).
115. Plumb, J.; Pigat, S.; Bompola, F.; Cushen, M.; Pinchen, H.; Nørby, E.; Astley, S.; Lyons, J.; Kiely, M.; Finglas, P. eBASIS (Bioactive Substances in Food Information Systems) and bioactive intakes: Major updates of the bioactive compound composition and beneficial bio effects database and the development of a probabilistic model to assess intakes in Europe. *Nutrients* **2017**, *9*, 320.
116. Lara-Cortes, E.; Osorio-Dıaz, P.; Jiménez-Aparicio, A.; Bautista-Bañios, S. Nutritional content, functional properties and conservation of edible flowers. *Review. Arch. Latinoam. Nutr.* **2013**, *63*, 197–208.
117. Matyjaszczyka, E.; Śmiechowsk, M. Edible flowers. Benefits and risks pertaining to their consumption. *Trends Food Sci. Tech*. **2019**, *91*, 670–674.

118. Kristanc, L.; Kreft, S. European medicinal and edible plants associated with subacute and chronic toxicity part I: Plants with carcinogenic, teratogenic and endocrine- disrupting effects. *Food Chem. Toxicol.* **2016**, *92*, 150–164.
119. Pinela, J.; Carvalho, A.M.; Ferreira, I.C.F.R. Wild edible plants: Nutritional and toxicological characteristics, retrieval strategies and importance for today's society. *Food Chem. Toxicol.* **2017**, *110*, 165–188.
120. Egebjerg, M.M.; Olesen, P.T.; Eriksen, F.D.; Ravn-Haren, G.; Bredsdorff, L.; Pilegaard, K. Are wild and cultivated flowers served in restaurants or sold by local producers in Denmark safe for the consumer? *Food Chem Toxicol.* **2018**, *120*, 129–142, doi:10.1016/j.fct.2018.07.007..
121. Klintschar, M.; Beham-Schimdt, C.; Radner, H.; Henning, G.; Roll, P. Colchicine poisoning by accidental ingestion of meadow saffron (*Colchicum autumnale*): Pathological and medicolegal aspects. *Forensic Sci. Int.* **1999**, *106*, 191–200, doi:10.1016/S0379-0738(99)00191-7.
122. Korkmaz, M.F.; Bostancı, M.; Onur, H.; Çağan, E. *Datura stramonium* poisoning: A case report and review of the literature. *Eur. Res. J.* **2019**, *5*, 186–188, doi:10.18621/eurj.39204.
123. Farkhondeh, T.; Kianmehr, M.; Kazemi, T.; Samarghandian, S.; Khazdair, M.R. Toxicity effects of *Nerium oleander*, basic and clinical evidence: A comprehensive review. *Hum. Exp. Toxicol.* **2020**, *39*, 773–784, doi:10.1177/0960327120901571.
124. Adler, L.S. The ecological significance of toxic nectar. *Oikos* **2001**, *91*, 409–420.
125. Rietjens, I.M.C.M., Martena, M.J., Boersma, M.G., Spiegelenberg, W., Alink, G.M. Molecular mechanisms of toxicity of important food-borne phytotoxins. *Mol. Nutr. Food Res.* **2005**, *49*, 131–158, doi:10.1002/mnfr.200400078.
126. Smith, R.L., Adams, T.B., Doull, J., Feron, V.J., Goodman, J.I., Marnett, L.J., Portoghese, P.S., Waddell, W.J., Wagner, B.M., Rogers, A.E., et al. Safety assessment of allylalkoxybenzene derivatives used as flavouring substances—Methyl eugenol and estragole. *Food Chem. Toxic.* **2002**, *40*, 851–870.
127. Grzeszczuk, M., Stefaniak, A., Meller, E., Wysocka, G. Mineral composition of some edible flowers. *J. Elem.* **2018**, *23*, 151–162, doi:10.5601/jelem.2017.22.2.1352.
128. Drava G, Iobbi V, Govaerts R, Minganti V, Copetta A, Ruffoni, B., Bisio, A. Trace elements in edible flowers from Italy: Further insights into health benefits and risks to consumers. *Molecules* **2020**, *25*, 2891, doi:10.3390/molecules25122891.
129. Lu, B.; Li, M.; Yin, R. Phytochemical Content, Health Benefits, and Toxicology of Common Edible Flowers: A Review (2000–2015). *Crit. Rev. Food Sci. Nutr.* **2016**, *56*, S130–S148, doi:10.1080/10408398.2015.1078276.
130. Akindahunsi, A.A.; Olaleye, M.T. Toxicological investigation of aqueous-methanolic extract of the calyces of Hibiscus sabdariffa L. *J. Ethnopharmacol.* **2003**, *89*, 161–164.
131. Fakeye, T.O.; Pal, A.; Bawankule, D.U.; Yadav, N.P.; Khanuja S.P.S. Toxic effects of oral administration of extracts of dried calyx of *Hibiscus sabdariffa* Linn. (*Malvaceae*). *Phytother. Res.* **2009**, *23*, 412–416.
132. Mahmoud, Y.I. Effect of extract of Hibiscus on the ultrastructure of the testis in adult mice. *Acta Histochem.* **2012**, *114*, 342–348.
133. De Arruda, A.; Cardoso, C.A.L.; Vieira, M.D.C.; Arena, A.C. Safety assessment of Hibiscus sabdariffa after maternal exposure on male reproductive parameters in rats. *Drug Chem. Toxicol.* **2015**, *39*, 1003938.
134. Mazzocchi, A.; Venter, C.; Maslin, K.; Agostoni, C. The role of nutritional aspects in food allergy: Prevention and management. *Nutrients*. **2017**, *9*, E850, doi:10.3390/nu9080850.
135. Costa, C.; Coimbra, A.; Vítor, A.; Aguiar, R.; Ferreira, A.L.; Todo-Bom, A. Food allergy-From food avoidance to active treatment. *Scand. J. Immunol.* **2020**, *91*, e12824, doi:10.1111/sji.12824.
136. Uthpala, T.G.G.; Navaratne, S.B.. *Acmella oleracea* plant; identification, applications and use as an emerging food source—Review. *Food Rev. Int.* **2020**, 1–16, doi:10.1080/87559129.2019.1709201.
137. Delort E, Jaquier A, Chapuis C, Rubin M, Starkenmann, C. Volatile composition of oyster leaf (*Mertensia maritima* (L.) Gray). *J Agr. Food Chem.* **2012**, 60, 11681–11690, doi:10.1021/jf303395q.
138. Najar, B.; Marchioni, I.; Ruffoni, B.; Copetta, A.; Pistelli, L.; Pistelli, L. Volatilomic analysis of four edible flowers from *Agastache* genus. *Molecules* **2019**, *24*, 4480, doi:10.3390/molecules24244480.
139. Marchioni, I.; Najar, B.; Ruffoni, B.; Copetta, A.; Pistelli La Pistelli, L.U. Bioactive compounds and aroma profile of some *Lamiaceae* edible flowers. *Plants* **2020**, *9*, 691, doi:10.3390/plants9060691.
140. Marchioni, I.; Pistelli, L.; Ferri, B.; Cioni, P.; Pistelli, L.; Ruffoni, B. Preliminary studies on edible saffron bio-residues during different post-harvest storages. *Bulg. Chem. Commun* **2019**, *51*, 131–136.

141. Bazzurro, V.; Milanese, M.; Gatta, E.; Bonifacino, T.; Diaspro, A.; Bonanno, G. Analisi in-vitro delle potenziali attivita' citotossiche su campioni rappresentativi di 40 estratti totali di fiori eduli. Available online: http://interregantea.eu/Poster.aspx (accessed on 18 September 2020).
142. Feo, F.; Martinez, J.; Martinez, A.; Galindo, P.A.; Cruz, A.; Garcia, R.; Guerra, F.; Palacios, R. Occupational allergy in saffron workers. *Allergy* **1997**, *52*, 633–641.