Check for updates

Received: 22 April 2024 | Revised: 16 August 2024 | Accepted: 18 August 2024

DOI: 10.1111/1541-4337.70015

COMPREHENSIVE REVIEW

Comprehensive REVIEWS in Food Science and Food Safety WILEY

Protein–phenolic interactions and reactions: Discrepancies, challenges, and opportunities

Helena Kieserling[1] | **Wouter J. C. de Bruijn**[2] | **Julia Keppler**[3] | **Jack Yang**[4] | **Sorel Tchewonpi Sagu**[5] | **Daniel Güterbock**[1] | **Harshadrai Rawel**[5] | **Karin Schwarz**[6] | **Jean-Paul Vincken**[2] | **Andreas Schieber**[7] | **Sascha Rohn**[1]

[1]Institute of Food Technology and Food Chemistry, Department of Food Chemistry and Analysis, Technische Universität Berlin, Berlin, Germany

[2]Laboratory of Food Chemistry, Wageningen University, Wageningen, The Netherlands

[3]Laboratory of Food Process Engineering, Wageningen University, Wageningen, The Netherlands

[4]Laboratory of Physics and Physical Chemistry of Foods, Wageningen University, Wageningen, The Netherlands

[5]Institute of Nutritional Science, University of Potsdam, Nuthetal, Germany

[6]Institute of Human Nutrition and Food Science, Division of Food Technology, Christian-Albrechts-Universität Kiel, Kiel, Germany

[7]Agricultural Faculty, Institute of Nutritional and Food Sciences, Molecular Food Technology, University of Bonn, Bonn, Germany

Correspondence
Sascha Rohn, Institute of Food Technology and Food Chemistry, Department of Food Chemistry and Analysis, Technische Universität Berlin, Gustav-Meyer-Allee 25, 13355 Berlin, Germany.
Email: rohn@tu-berlin.de

Abstract

Although noncovalent interactions and covalent reactions between phenolic compounds and proteins have been investigated across diverse scientific disciplines, a comprehensive understanding and identification of their products remain elusive. This review will initially outline the chemical framework and, subsequently, delve into unresolved or debated chemical and functional food-related implications, as well as forthcoming challenges in this topic. The primary objective is to elucidate the multiple aspects of protein–phenolic interactions and reactions, along with the underlying overwhelming dynamics and possibilities of follow-up reactions and potential crosslinking between proteins and phenolic compounds. The resulting products are challenging to identify and characterize analytically, as interactions and reactions occur concurrently, mutually influencing each other. Moreover, they are being modulated by various conditions such as the reaction parameters and, obviously, the chemical structure. Additionally, this review delineates the resulting discrepancies and challenges of properties and attributes such as color, taste, foaming, emulsion and gel formation, as well as effects on protein digestibility and allergenicity. Ultimately, this review is an opinion paper of a group of experts, dealing with these challenges for quite a while and aiming at equipping researchers with a critical and systematic approach to address current research gaps concerning protein–phenolic interactions and reactions.

KEYWORDS
covalent reactions, crosslinking, physiological properties, protein–phenolic interactions, technological properties

This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.
© 2024 The Author(s). *Comprehensive Reviews in Food Science and Food Safety* published by Wiley Periodicals LLC on behalf of Institute of Food Technologists.

1 | INTRODUCTION

It has been assumed that plant phenolic compounds (PCs) positively affect human health, with a long list of suggested bioactivities (Fraga et al., 2019; Scalbert et al., 2007) and also adverse effects (Lambert et al., 2007; Sinha et al., 2022). The mechanisms of the bioactivity of PCs are not fully understood yet, mainly due to the multitude of physiological and biochemical aspects that need to be considered, such as uptake, digestibility, absorption throughout the gastrointestinal tract, systemic metabolism, bioaccessibility in tissues, and excretion rate. Although these factors are intensively studied, contradictory results are described from study to study. Moreover, PCs can interact with proteins before even being consumed, thereby greatly diversifying their structures and properties.

The complexity of the effects of protein–phenolic interactions may be attributed to the various composition of foods. In food, several different constituents, including PCs, bear the potential of interacting noncovalently or reacting covalently with each other. The chemistry behind these interactions is still too complex to reveal the exact chemical structure of specific protein–phenolic interaction and reaction products. Due to the high structural diversity of the products, many of which are of higher molecular weight, traditional analytical methods, for example, chromatography, encounter challenges in achieving sufficient separation. Consequently, these interaction and reaction mixtures contain many unrevealed or unknown compounds. However, not all proteins and PCs react completely with each other, but it is rather the case that certain chemical (sub)structures are more or less susceptible to interactions or reactions. Basic chemical mechanisms induce a significant change with subsequent consequences for physicochemical properties, technological properties, and physiology.

More and more researchers are characterizing interactions between PCs and other food ingredients. When looking at food composition in the context of foods rich in PCs, the main interaction partners are proteins, as they provide several structural features for interacting or reacting with PCs. Besides, the high PC concentration and the low protein:PC ratio increase the interaction probability. Proteins can interact noncovalently but can also form covalent reaction products with PCs. The preference for noncovalent or covalent interaction is highly dependent on the conditions, as well as on the initial properties of the PCs and the proteins. Main challenges for the topic presented herein are the large structural diversity in small as well as polymeric PCs, the large variety of proteins, and the many different conditions under which both compound classes are forced to interact or react with each other.

Protein–phenolic interactions and reaction products affect the sensorial, technological, and physiological properties of the corresponding foods. In this context, one must only think of polymeric reaction products that affect the food's color and solubility, resulting in unrevealed nutritional consequences. However, there are also possibilities of desirable interactions for beneficially modifying foods' properties. In times of protein transition toward increasingly plant protein-rich ingredients, recipes, and products, the topic of protein–phenolic interactions and reactions becomes progressively a priority, as plant produce is generally rich in PCs.

Although most of the aspects mentioned so far seem to be easy to handle, many factors still make the topic of protein–phenolic interactions and reactions quite challenging. The different sections of this review present both the chemical backgrounds and the functional consequences of protein–phenolic interactions, as well as the many challenges that remain to be overcome. Building upon the reviews of recent years, for example, Shahidi and Dissanayaka (2023) and Tarahi et al. (2024), both the complexity of the different aspects and their close connection with each other are presented. So far, there is a lack of systematic studies that employ consistent, widely accepted analytical methods to identify the full range of protein–phenolic interaction and reaction products, such as chromatographic (e.g., LC–MS) and spectroscopic (e.g., FTIR) analyses or molecular dynamic simulations. Thus, this overview intends to summarize relevant phenomena, to face challenges more systematically, and to apply this knowledge to develop more stable, healthy, and sustainable foods.

2 | UNRAVELING THE MECHANISMS OF PROTEIN–PHENOLIC INTERACTIONS AND REACTIONS

Interactions between proteins and both "non-oxidizable" PCs (Figure 1, No. 1) and "oxidizable" PCs (No. 2) comprise reversible binding, resulting in noncovalent interaction products (Nos. 3 & 4) or irreversible binding by covalent bonds. The latter is referred to as protein–phenolic "reaction products" or "adducts" (No. 5). Consequently, in this review, the term "interaction" refers to noncovalent binding, whereas "reaction" denotes covalent binding between PCs and proteins. Terms like "complexation" or "conjugation" are often used in the literature, but these could be misleading, as complexation might refer to specific forms of interactions, while "conjugation" is a term that is often used in the context of metabolism.

The occurrence and the preference for a specific type of protein–phenolic interaction or reaction strongly depend

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

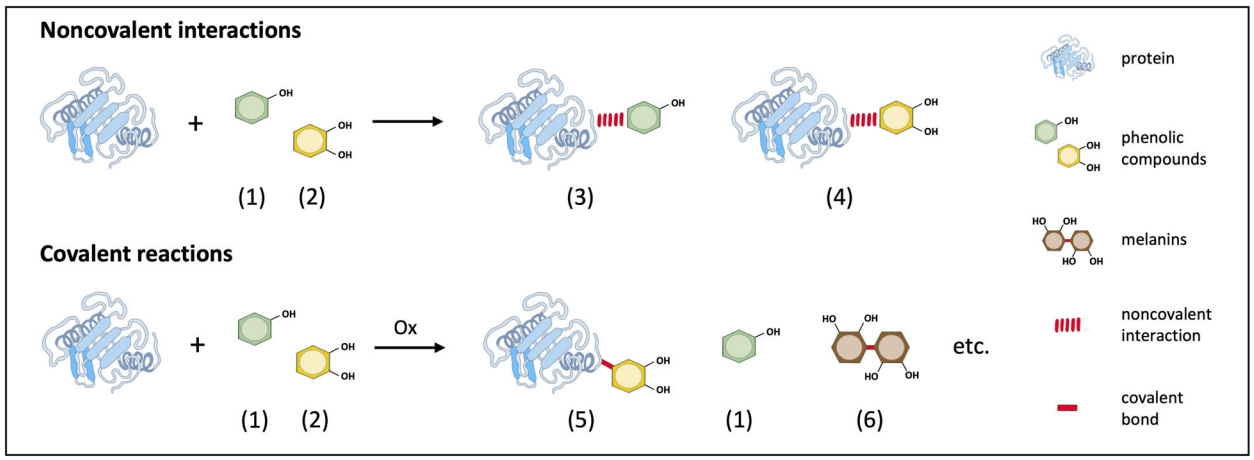

FIGURE 1 Schematic overview of noncovalent interactions and covalent reactions between proteins and phenolic compounds (PCs), leading to initial interaction and reaction products. The PCs are simplified into two groups: (1) monohydroxy PCs, which represent PCs capable of engaging in noncovalent interactions and (2) *o*-dihydroxy PCs, which represent PCs capable of both noncovalent interactions and forming covalent bonds through *o*-quinone formation. Typically, such PCs exhibit additional structural features, which are not depicted here. Potential protein structural changes are neglected for the sake of simplicity.

on various factors, including the physicochemical conditions as well as the structural features of the PCs and the proteins. The following subsections describe the interaction and reaction mechanisms (Subsection 2.1), followed by a more in-depth discussion of the effect of various factors on both the formation of noncovalent and covalent products (Subsection 2.2). Finally, the current knowledge gaps related to protein–phenolic interactions in food are discussed (Subsection 2.3).

2.1 | Possibilities of interaction and reaction: Noncovalent versus covalent

Figure 1 illustrates that proteins and PCs (Nos. 1 & 2) can form either noncovalent interaction products (Nos. 3 & 4) or react to adducts (Nos. 5 & 6) under oxidative conditions. The mechanistic principles of both interaction and reaction are described in the following subsections.

2.1.1 | Basics of noncovalent interactions between proteins and phenolic compounds

Noncovalent interactions between proteins and PCs are mainly stabilized by a combination of reversible hydrophilic and hydrophobic interactions. As depicted in Figure 2, these interactions mainly include hydrogen bonds, ionic or electrostatic interactions, and hydrophobic stacking interactions (π–π and CH–π). In addition, van der Waals' forces and nonstacking hydrophobic interactions with nonaromatic hydrophobic protein side chains of certain amino acids (e.g., isoleucine, leucine, methionine, cysteine) generally have a relatively small contribution to the interactions (Condict et al., 2019; Dai et al., 2022; Richard et al., 2005).

Hydrogen bonds can be formed between hydroxyl groups of the aromatic ring and carbonyl or amino groups of the protein backbone (No. 7), as well as amino, hydroxyl, thiol, or carbonyl groups in the protein side chains (Croft & Foley, 2008). Ionic/electrostatic interactions occur between negative charges of phenolate anions or other deprotonated moieties (e.g., carboxylate anions) of the PCs and positive charges in protein side chains (No. 8) (Le Bourvellec & Renard, 2012). These charge-driven interactions are strongly dependent on the physicochemical conditions of the system, primarily the pH, which affects the presence and exposure of positively charged protein side chains (i.e., arginine, histidine, and lysine). Additionally, the pK_a of a PC is relevant for a charge-driven interaction. For example, phenolic acids with a low pK_a, such as ferulic acid or chlorogenic acid, are negatively charged under neutral pH conditions (Li, Li et al., 2020), which results in charge-driven interaction with positively charged amino acid residues (cf. Subsection 2.2). Hydrophobic stacking occurs via "face-to-face" overlapping between the π-orbitals of the aromatic ring of the PCs and the π-orbitals of aromatic groups in the corresponding protein side chains (No. 9; i.e., histidine, phenylalanine, tryptophan, and tyrosine) or via perpendicular or angular stacking of the π-orbitals of the phenolic ring with the H_α of the amino acid proline (No. 10) (Baxter et al., 1997; Croft & Foley, 2008; Zondlo, 2012). Intriguingly, particularly the latter interactions involving proline seem to occur quite frequently, as CH–π stacking is commonly reinforced by hydrogen bonds (cf. Figure 2).

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

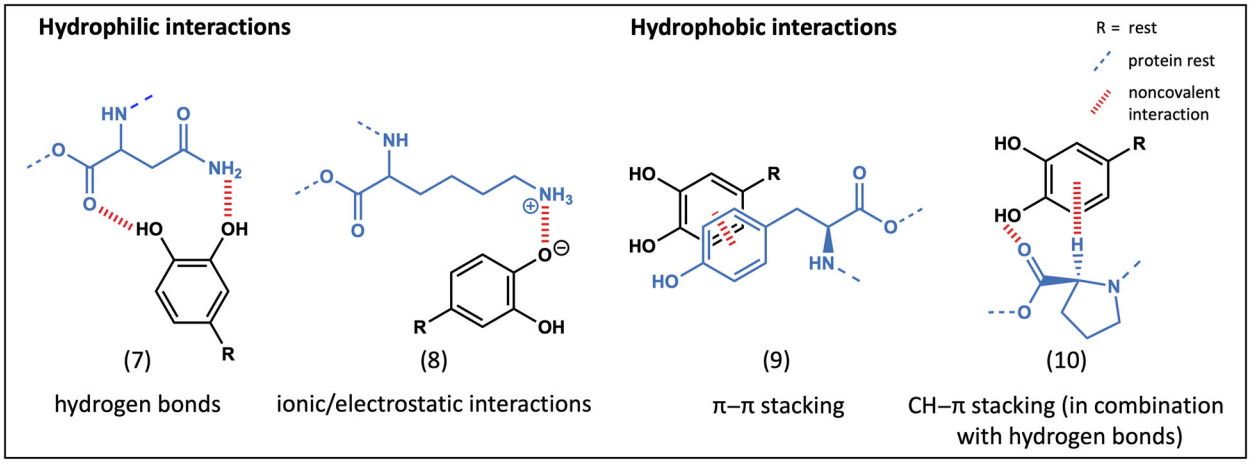

FIGURE 2 Overview of the main noncovalent interactions between proteins and phenolic compounds (PCs). PCs are shown in black, proteins in blue, and interactions in red.

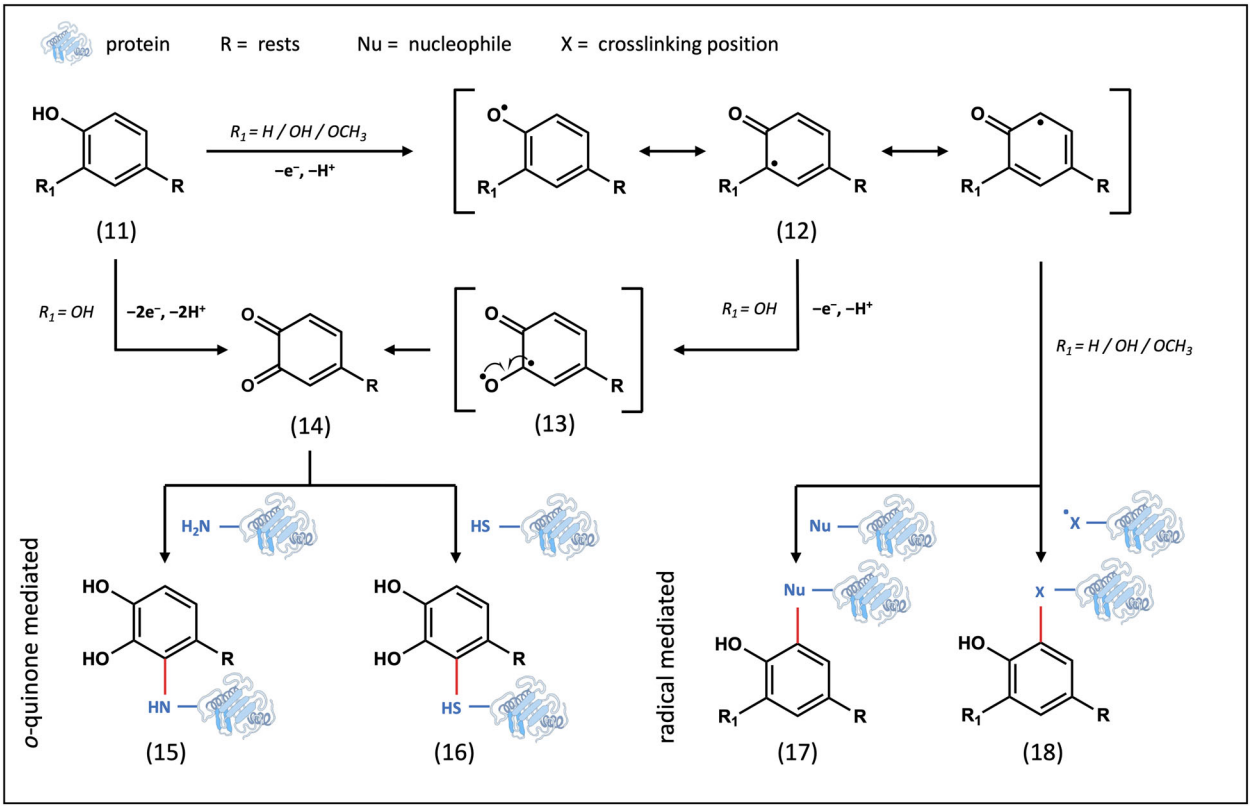

FIGURE 3 Overview of oxidation reactions underlying covalent protein–phenolic reactions. The o-quinone-mediated bond formation is drawn in an arbitrary position on the aromatic ring. In practice, the position is dependent on the aromatic substituents. Potential protein structural changes are neglected for the sake of simplicity.

2.1.2 | Basics of covalent bond formation between proteins and phenolic compounds

Covalent protein–phenolic bond formation occurs primarily via oxidation of the PCs, followed by coupling reactions (Figure 3). The most prominent mechanism of protein–phenolic reactions is a MICHAEL addition of nucleophilic groups from the protein side chains to electron-deficient *o*- or *p*-quinones resulting from the oxidation of PCs.

The formation of quinones can proceed enzymatically or autoxidatively. Enzymatic oxidation of 1,2-hydroxybenzene moieties (No. 11) to *o*-quinones (No.

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

14) is commonly catalyzed by polyphenoloxidases (e.g., tyrosinases and catecholoxidases) or related oxidases (e.g., peroxidases). For instance, tyrosinase is responsible for two successive reactions in the presence of molecular oxygen: (I) the hydroxylation of monophenols to form *o*-diphenols (monophenolase activity) and (II) the oxidation of *o*-diphenols to *o*-quinones (diphenolase activity). Autoxidation mainly occurs at alkaline pH values or in the presence of oxidizing agents (e.g., transition metals).

Typically, quinones react with themselves to form brown melanin pigments (Figure 1, No. 6), which exhibit a complex chemical structure comprising numerous phenolic moieties. They are often not fully elucidated (Cao et al., 2021; Friedman, 1996; Galeb et al., 2023). Moreover, highly reactive quinones can participate in reactions with various other reaction partners (Bittner, 2006; Rohn, 2014; Schieber, 2018). Here, the focus is on the addition of quinones with nucleophilic groups of the protein side chains, which leads to covalent protein–phenolic reaction products (Nos. 15 & 16). Addition primarily occurs at side chains that contain amino (i.e., lysine) or thiol (i.e., cysteine) groups (Drucker et al., 2023). As recently confirmed by Lund (2021), reaction rates with thiol groups are up to 10^5 times higher than those observed with amino groups. Hence, proteins with a higher number of free thiol groups (e.g., bovine serum albumin) typically react faster with quinones than proteins without free thiol groups (e.g., α-lactalbumin). Additionally, the reaction of quinones with various other amino acids in proteins, including guanidino (i.e., arginine), indole (i.e., tryptophan), amide (i.e., asparagine, glutamine), imidazole (i.e., histidine), or aromatic (i.e., tyrosine, phenylalanine) groups, has also been reported (Bittner, 2006; Lund, 2021; Prigent et al., 2003; Rawel et al., 2001). However, these amino acids have mostly been studied individually rather than systematically in a comparative manner, particularly regarding their structural features, leading to ongoing controversy regarding their reactivity with proteins.

Besides *o*- or *p*-quinone-mediated protein–phenolic reactions, radical-mediated coupling has also been reported. Radical-mediated coupling proceeds from semiquinone-type radicals (No. 12) that are formed upon oxidation of phenolic moieties. Semiquinones can be generated enzymatically or autoxidatively from hydroxybenzenes (e.g., coumaric acid), *m*-dihydroxybenzenes (e.g., resorcinol), and 1-methoxy-2-hydroxybenzene moieties (e.g., ferulic acid), but of course also *o*- or *p*-dihydroxybenzene-containing derivatives (e.g., caffeic acid). However, the latter structural feature undergoes relatively fast subsequent oxidation to the *o*- or *p*-quinones (e.g., Figure 3, Nos. 13 & 14). The semiquinone radicals from the other structural features are stable, being able to react subsequently with nucleophilic groups or with a radical from an oxidized protein to form a covalent bond (Nos. 17 & 18) (Rawel & Rohn, 2010; Rohn, 2014; Zhang et al., 2021).

Most dominantly, during the MICHAEL addition, hydroxyls are recovered, and subsequent oxidation of the protein-bound PCs to their quinones (or semiquinones) can again induce the addition of further PC molecules or even covalent protein crosslinking (cf. Sections 3 and 4). The covalent bonds formed via nucleophilic addition to quinones or semiquinones are irreversible, because they lead to stable conjugated or even aromatic systems (Schieber, 2018), whereas noncovalent interactions are more or less reversible. A further discussion of the reaction dynamics and the nature of covalent crosslinking is provided in Sections 3 and 4.

2.2 | Factors affecting noncovalent interaction and covalent reaction between proteins and phenolic compounds

It should be emphasized that interactions of PCs and proteins and the formation of reaction products are often discussed separately. However, they co-exist and are collectively responsible for foods' functionality.

Still, the prevalent mechanism underlying protein–phenolic interactions and the extent of protein modification strongly depend on the physicochemical conditions of the system, the chemical–structural properties of the PCs, and the structural properties of the proteins. The influence of these factors is summarized in Tables 1 and 2.

2.2.1 | Effect of physicochemical conditions

The main physicochemical conditions affecting protein–phenolic interactions are the pH value, temperature, and ionic strength. Briefly, noncovalent protein–phenolic interactions are typically preferred at acidic conditions and temperatures that affect protein structure (Zhang et al., 2021).

Ionic strength can either negatively or positively influence the binding affinity (Le Bourvellec & Renard, 2012; Prigent et al., 2003; Rawel et al., 2005), potentially by neutralizing charges and preventing charge complexes between PCs and proteins. Varying findings on the effect of temperature have been reported. For example, Prigent et al. (2003) reported that the number of 5-*O*-caffeoylquinic acid molecules bound to bovine serum albumin decreased with increasing temperature from 25 to 60°C, whereas Tsai and She (2006) reported that binding of various PCs to superoxide dismutase increased with temperature. Hofmann et al. (2006) reported that the temperature increased

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

TABLE 1 Factors influencing noncovalent interaction of proteins and phenolic compounds (PCs) (modified from Keppler et al., 2020; Le Bourvellec & Renard, 2012; Zhang et al., 2021).

Factor	Effect on noncovalent interaction products
pH	Strongest at acidic conditions (pH ≈ 3)
Temperature	Some discrepancies in literature, generally highest binding affinity expected around 25°C Heat-induced protein denaturation → binding sites ↑ Heat-induced protein aggregation → binding sites ↓
Moisture content	Unknown
Ionic strength	Discrepancies in literature, ionic strength can have a positive or negative effect depending on noncovalent interactions involved
PC structure	↑ molecular size (# of phenolic moieties) → binding affinity ↑ ↑ flexibility → binding affinity ↑ Common functional groups (—OH, —OCH_3, and —COOH) affect noncovalent interaction Flavonoid-specific binding affinity affected by hydrophobicity, galloylation and glycosylation, and C-ring structure
Protein structure	Random coil or helical > globular ↑ proline content → binding affinity ↑ ↑ flexibility → binding affinity ↑ Exposure and accessibility of relevant residues or regions
Ratio phenol/protein	Low ratio: small, soluble complexes Medium ratio: noncovalent aggregation of proteins via multidentate complexation of PCs leads to larger complexes with decreased solubility High ratio: protein aggregation and precipitation
Presence of oxygen	No direct effect
Presence of oxidants	No direct effect, indirectly might lead to a shift toward covalent reactions
Presence of anti-oxidants	No direct effect, indirectly might lead to a shift toward noncovalent interactions

TABLE 2 Factors influencing covalent reaction of proteins and phenolic compounds (modified from Keppler et al., 2020; Le Bourvellec & Renard, 2012; Zhang et al., 2021).

Factor	Effect on covalent interaction products
pH	Nonenzymatic requires alkaline conditions (pH >7) Enzymatic: Maximum at enzyme optimum (pH 5–7), enzymes inhibited by acid (pH <4) Deprotonation of NH_3^+/SH at high pH → nucleophilicity ↑ → degree of covalent binding ↑
Temperature (T)	Nonenzymatic: ↑ T → rate of autoxidation ↑ Enzymatic: Maximum at enzyme optimum (typically 25–35°C), enzyme inactivation >60°C Heat-induced protein denaturation → binding sites ↑ Heat-induced protein aggregation → binding sites ↓
Moisture content	Possibly promoted by high moisture content
Ionic strength (IS)	Unknown
PC structure	Enzymatic: Substrate specificity of the enzyme: chlorogenic acid, caffeic acid, and catechins are examples of preferred substrates Preferable via *o*-quinone formation from 1,2-dihydroxybenzene moiety ↓ Redox potential of the phenol → oxidation ↑
Protein structure	Accessibility and number of nucleophilic amino acid side chains → ↑ degree of covalent binding
Ratio phenol/protein	↑ ratio → degree of covalent binding ↑
Presence of oxygen	Oxygen is required for *o*-quinone or semiquinone generation via oxidation
Presence of oxidants	Nonenzymatic: Promotes oxidation Enzymatic: Not required
Presence of anti-oxidants	Inhibits *o*-quinone and semiquinone reactions with proteins

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

the interaction of hydrolyzable tannins with bovine serum albumin, but it did not affect the binding of smaller procyanidins. It seems that the increase in temperature lowers the bond strength of the hydrogen bonds, while hydrophobic interactions increase (Prigent et al., 2009). Therefore, the discrepancies between the different studies can likely be explained by the different structural characteristics of the PCs investigated (polar vs. nonpolar, small vs. large), resulting in distinct binding modes to the different protein types. At the same time, however, protein structures are also affected by a mild temperature increase, because secondary and tertiary structure elements undergo a certain degree of refolding and are stabilized by hydrogen bonds and hydrophobic interactions. Even minor changes in protein structure can affect the interaction with various PCs. Thus, complex interaction dynamics are at play (cf. Section 3).

The factors affecting covalent protein–phenolic reactions are mainly the pH, temperature, and particularly oxidative conditions. The extent of enzymatic oxidation, resulting in protein–phenolic reaction, depends on the conditions that determine polyphenoloxidase activity. Optimal conditions for polyphenoloxidase activity are typically slightly acidic to neutral pH values and temperatures from 25 to 35°C (Queiroz et al., 2008). At acidic conditions (pH <4), the enzyme activity is reversibly inhibited, whereas at higher temperatures (>60°C), denaturation, affecting the enzyme activity, occurs. Autoxidation happens readily under alkaline conditions (pH >7) and increases the oxidation rate at higher temperatures. High moisture content promotes both ways of phenolic oxidation (i.e., autoxidation and enzymatic oxidation) and thereby the formation of protein–phenolic reaction products (Keppler et al., 2020). Furthermore, more alkaline pH values (≥9) promote the reaction indirectly by deprotonation of thiol groups, which enhances their nucleophilicity and thereby their reactivity toward *o*-quinones and semiquinones (Bulaj et al., 1998; Keppler et al., 2020).

As indicated in Tables 1 and 2, the discrepancies in literature regarding the effect of various conditions on noncovalent protein–phenolic interactions are likely due to a variation in experimental setups and a strong interplay between physicochemical conditions and intrinsic factors. As already mentioned, noncovalent interaction and covalent reaction can occur simultaneously. It is therefore complex to isolate single effects caused solely by noncovalent interaction or covalent binding, as many studies do not clearly exclude the other mode of binding. Furthermore, the samples are not static, especially when studying noncovalent interactions. That means that autoxidation and subsequent covalent attachment of PCs can occur even at neutral pH, albeit at significantly lower reaction rates than at alkaline pH, depending on the PC structure and PC concentration (Jia et al., 2022). Slight shifts in the ratio from noncovalent toward covalent modifications occur with time and can affect the experimental outcome. Another major challenge that adds to the discrepancies about noncovalent binding is that these are hard to measure, especially when the reactivity is of low affinity (Keppler et al., 2014; Schild et al., 2023).

Additionally, compared to some findings already mentioned, multiple studies claim opposite results regarding the interaction of the same whey protein with epicatechin or with gallic acid (Kanakis et al., 2011; Li, Dai, et al., 2020; Nucara et al., 2013; Riihimäki et al., 2008), even at neutral pH and room temperature. Besides variations in methods, the protein can also be responsible for observed differences in interacting with the same PC. For commercial ultrapure protein standards, the glycation level, nativity, and oxidation level vary batchwise, so that the protein status can affect the observed interaction kinetics (Subsection 2.2.2), together with the structure of the PC (Subsection 2.2.3).

2.2.2 | Effect of structural properties of the protein

In addition to the chemical structure of the PCs, the proteins' structural properties strongly affect the extent and the nature of protein–phenolic interactions, as well as the interaction dynamics. It should be noted that most of this information is based on research on interactions of model PCs with model proteins and only a few "real" food-related protein systems like milk or proline-rich salivary proteins.

All protein structure elements—that is, primary, secondary, tertiary, and quaternary—affect noncovalent interaction with PCs. Among the listed amino acids in Subsection 2.1.1, proline residues are the most prominent examples mentioned when trying to explain noncovalent interactions with proteins. The first studies used proline-rich salivary proteins to check interactions and try to explain the astringent taste of PCs (Bacon & Rhodes, 1998; Baxter et al., 1997). Proline residues strongly enhance the PC binding capacity, forcing the protein to adopt a more irregular folded or extended structure (Le Bourvellec & Renard, 2012; Richard et al., 2005). In fact, the more unfolded the protein secondary and tertiary structures are, the higher the proteins' structural flexibility, providing a high accessibility of binding sites and hence, promoting phenolic interactions.

Consequently, some highly ordered β-sheet-rich globular proteins (e.g., β-lactoglobulin) typically interact with PCs via binding at a limited number of specific regions in the protein. Moreover, interactions between PCs and highly ordered fibrils of, for example, animal-derived proteins have recently garnered increasing attention (Li et al.,

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

2021; Xu et al., 2022; Yan et al., 2024). Globular proteins (e.g., bovine serum albumin) and most random coil- or α-helix-dominated proteins (e.g., β-caseins, gelatin) tend to possess a high number of binding regions (Baxter et al., 1997; Bohin et al., 2012; Kanakis et al., 2011; Li, Li et al., 2020). Such an effect was also observed for different genetic variants of β-lactoglobulin; β-lactoglobulin A has a more flexible structure than β-lactoglobulin C. The latter has two amino acid mutations that result in an internal salt bridge. This lowered the number of binding sites for epigallocatechin gallate from two in β-lactoglobulin A to one in β-lactoglobulin C. Temperature-induced denaturation of both proteins resulted in unfolding and increased the number of binding sites to two for both proteins (Keppler et al., 2014). This demonstrates that minor changes by only two amino acids in the otherwise same protein can already significantly affect the interaction with the same PC. Moreover, it is expected that proteins with a rigid tertiary structure (often due to a high number of disulfide bonds) and with a highly multimeric quaternary structure (e.g., oligomeric proteins) could have lowered accessibility of the binding sites, which limit their interaction possibilities with PCs.

Likewise, for the formation of covalent protein–phenolic reaction products, the main determining factors in the protein structure are the accessibility and the number of nucleophilic protein side chains such as lysine and cysteine.

All interactions and reactions influence each other because both can induce structural changes in the protein, which can either reveal additional binding sites or bury existing ones, influencing the interaction and reaction dynamics. This complicates the characterization of binding sites and elucidation of the underlying mechanisms. However, in the literature, there is not yet a systematic answer available to the following questions: Do PC-induced structural changes occur in the first place? When they do, under which conditions and to which extent? As illustrated in Figure 4, there might be no structural changes at all (Figure 4, No. 19). In fact, some studies describe a loss in ordered secondary structure elements like α-helix and β-sheet and an increase in irregular random coil structures (Figure 4, No. 20) (e.g., for mixtures of β-lactoglobulin with chlorogenic acid) (Jia et al., 2017). Other studies with similar substances state an α-helix- to β-sheet transition (No. 21) (Sui et al., 2018). Moreover, some studies even suggest an increase in ordered and a decrease in irregular structures (No. 22) (Li, Dai, et al., 2020; Prigent et al., 2003).

Even though these discrepancies can partly be traced back to the previously mentioned difficulties in separating noncovalent and covalent as well as protein effects, most studies have in common that the structural elements change because of noncovalent or covalent protein modification, indicating a change in binding site accessibility after adding PCs. For noncovalent interactions with different PCs in tea, the structural changes observed for the protein correlated positively with binding affinity (Schild et al., 2023). Furthermore, structural changes are becoming more pronounced as more PCs are bound, for both noncovalent and covalent binding (Jia et al., 2017). Consequently, the interactions and reactions influence the dynamics of protein structural changes, as illustrated in Figure 5 (Nos. 20a–20c). However, proteins with a more rigid secondary and tertiary structure potentially retain their native structure to a larger extent after interaction with PCs than less stable proteins. It would be highly interesting to elucidate the dynamics between noncovalent and covalent modifications and to which level these influence each other. For example, one could surmise that PCs first bind noncovalently to a protein, change the secondary or tertiary structure, and thereby enhance the accessibility of free thiol or amino groups for covalent modifications. Conversely, opposite effects of negative cooperativity are also possible, meaning that noncovalent modification can increase the rigidity of the protein and thereby block binding sites. Such interactions would be promising subjects of future studies because they can be used for the targeted induction or prevention of excessive protein–phenolic interactions or crosslinks (cf. Section 3).

2.2.3 | Effect of chemical–structural properties of the phenolic compound

Protein–phenolic interaction is strongly influenced by the chemical structure of PCs. Generally, noncovalent interaction with proteins is stronger for PCs that contain multiple phenolic moieties. For single-subunit small molecules, substituents at the aromatic ring and other structural features (i.e., residual groups R in Figure 3) affect binding affinities (Bohin et al., 2012; de Paula Rezende et al., 2020; Jin et al., 2012; Kanakis et al., 2011; Zhang et al., 2021). For example, Jin et al. (2012) reported that steric hindrance of the aromatic methoxyl groups in ferulic acid and sinapic acid diminished the binding affinity toward bovine serum albumin compared to chlorogenic acid and caffeic acid, which do not contain methoxyl groups. Similarly, the interaction of epigallocatechin gallate with bovine serum albumin occurred via π–π stacking with additional stabilization by three hydrogen bonds, whereas the interaction of gallic acid exclusively occurred electrostatically (Skrt et al., 2012).

For single-subunit, but large flavonoid molecules, additional phenolic moieties, such as those present in

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

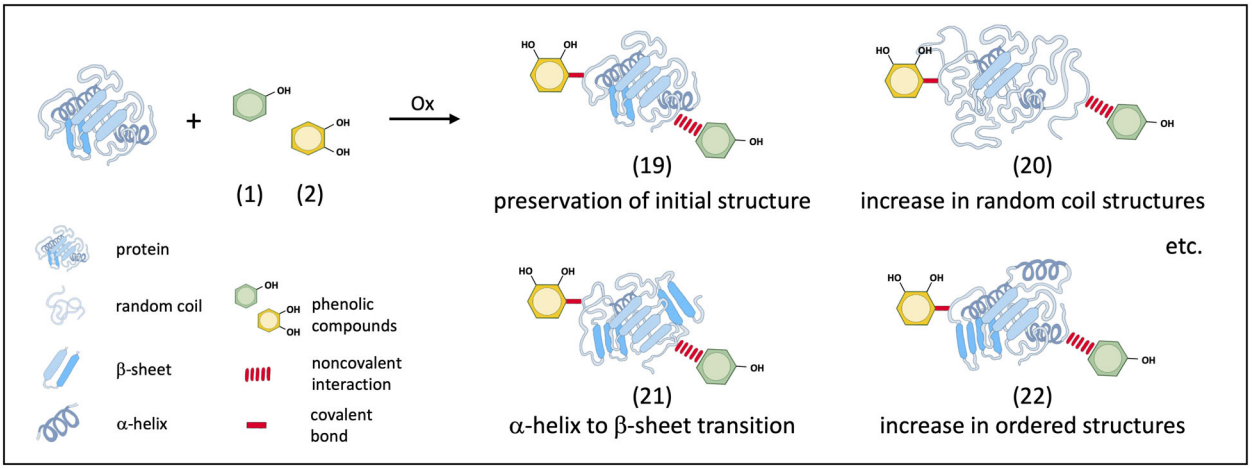

FIGURE 4 Potential structural modifications of proteins induced by interaction or reaction with phenolic compounds. Different possibilities of interaction and reaction as well as evaluation of preferred pathways are neglected for the sake of simplicity.

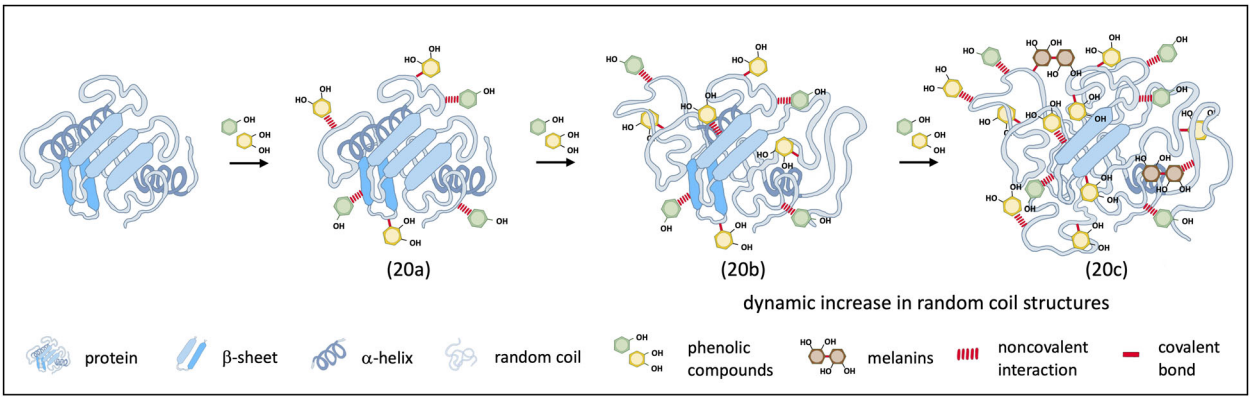

FIGURE 5 Potential dynamics of protein structural changes, like the increase in irregular random coil structures, induced by interaction and reaction with phenolic compounds.

galloylated catechins, increased binding affinity (Poncet-Legrand et al., 2006). Additionally, the C-ring structure and overall hydrophobicity of the single flavonoid molecules affected their noncovalent interaction with different proteins (Bohin et al., 2012). Interaction of proteins with PCs that have high binding affinity can lead to extensive coating of the protein, which may in turn result in noncovalent aggregation and protein precipitation (cf. Section 4). This behavior is mainly observed for di-, oligo-, or even polymeric PCs such as hydrolyzable and condensed tannins, which typically form more pronounced interactions with proteins than simple monomeric PC molecules. A higher number of structural features, such as more hydroxyl groups, more aromatic rings, and larger areas of hydrophobic or hydrophilic character, increases the possibility of interactions with proteins and the ability to form multidentate complexes with further complexation agents (Baxter et al., 1997; Le Bourvellec & Renard, 2012; Prigent et al., 2003). The term of complexation describes in this case a specific form of noncovalent interaction that involves further interaction partners such as metal ions (Bijlsma et al., 2020). Thus, with an increasing degree of molecular size of PCs, their ability to aggregate and precipitate proteins is enhanced.

As described in Subsection 2.1.2, covalent bond formation is mainly induced by the preceding generation of *o*- or *p*-quinones. For this reason, the presence of a 1,2- or a 1,4-dihydroxybenzene moiety of a PC is a prerequisite. Nevertheless, covalent bond formation between 1-methoxy-2-hydroxybenze moieties (e.g., ferulic acid) via semiquinone-type radicals is also possible (Condict et al., 2019; Kallinich et al., 2018). For autoxidation, an essential factor determining whether PC oxidation will occur is their redox potential. Thus, PCs with low redox potentials are more reactive regarding covalent protein–phenolic reactions. For initial enzymatic oxidation, the substrate

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

specificity of the enzyme is a further main determinant for quinone formation. Substrate acceptance depends on the other structural features (i.e., residual group R in Figure 3) besides the 1,2-dihydroxybenzene moiety. Chlorogenic acid, caffeic acid, and catechins are examples of reactive substrates that are, therefore, widely used for covalent protein–phenolic modification (Keppler et al., 2020; Ozdal et al., 2013). Moreover, quinone-mediated condensation of monomeric PCs (e.g., caffeic acid) to their corresponding oligomers or polymers ("melanins") can increase their reactivity toward amino acid side chains (Prigent et al., 2009) and enable the formation of benzacridines (Schieber, 2018) (cf. Sections 3 and 5).

There are some controversies about the effect of the structural properties of PCs on protein–phenolic interactions. While the overall topic of protein–phenolic interactions and reactions faces challenges due to the complexity of the interaction and reaction dynamics, certain studies have approached the topic systematically and comparatively, typically using the same protein and reaction conditions while varying only the chemical structures of the PCs. The observed effects are quite general and robust, even for different proteins. However, when multiple PCs are present simultaneously, the complexity increases, leading to competition for binding sites (affecting noncovalent interactions and covalent reactions), positive or negative cooperativity of the proteins, and different oxidation kinetics (primarily affecting covalent interactions). Studies addressing these more complex scenarios are currently scarce but would be highly relevant for understanding increasingly intricate systems.

2.3 | Determining protein–phenolic interactions in food is a challenge

From the preceding subsections, one could conclude that protein–phenolic interactions and reactions occur in an ordered, controllable, and predictable manner. This is not typically the case, as this is only true—when at all—under certain model conditions. When it comes to "real" complex food systems, there are many proteins and PCs that have not yet been investigated as starting materials for interactions and reactions in their full range. For instance, only a limited number of studies investigated the interactions of plant proteins with (endogenous) plant PCs. A vast majority of current knowledge on protein–phenolic interactions is based on studies performed with pure or mixed milk proteins and with other animal proteins such as gelatines and serum albumins. However, with current trends shifting to plant-based foods, the importance of plant proteins' interaction and reaction with PCs increases extensively. Consequently, this means that the relevance of protein–phenolic interactions and reactions is increasing even more, as plant protein fractions typically also contain PCs, depending on their degree of purity (e.g., meals vs. concentrates vs. isolates). The phenomena of decreased protein solubilities and adverse protein coloration as observed during alkaline protein isolation have been established for a long time (Sabir et al., 1974; Sosulski, 1979; Wildermuth et al., 2016).

Furthermore, in complex food systems, the composition and the protein:PC ratio vary, for example, in protein- or PC-rich foods. As indicated in Tables 1 and 2, the protein:PC ratio strongly influences protein–phenolic interactions and reactions and has also recently been demonstrated by Jia et al. (2022) and reviewed by Shahidi and Dissanayaka (2023) (cf. Subsections 2.1 and 2.2).

Moreover, one can deduce from the previous subsections that processing foods containing proteins and PCs can induce desirable or undesirable protein–phenolic interactions. In addition, other compounds present may interfere with or affect protein–phenolic interactions and reactions as well, for example, by competitive binding (e.g., glycation) and involvement in redox reactions. Finally, the pH value of the food strongly determines the interaction of PCs and proteins as well (for instance, alkaline egg proteins vs. acidic whey) (Keppler et al., 2020). So far, there is no systematic insight into the effects and interplay of processing and storage conditions on protein–phenolic interactions and reactions. Additionally, it is not possible to predict the outcome, especially the reaction dynamics, of whether the protein reacts with PCs first or whether the PCs react with each other to form melanins before interacting with the protein (cf. Section 3). These interactions and reactions can have a pronounced influence on protein functionality and hence on food quality (Zhang et al., 2021). As highlighted in Subsections 2.1 and 2.2, there are already many controversies about binding effects and structural changes in model systems. Of course, such discrepancies are further multiplied in complex systems and will also be reflected in the reported changes in protein functionality (cf. Section 5), because proteins' functionalities are a consequence of their structure.

3 | INTERACTION AND REACTION DYNAMICS RESULT IN COMPLEX MIXTURES OF PRODUCTS

As already indicated in Section 2, it is difficult to separate individual phenomena solely based on covalent or noncovalent modifications. Therefore, in this section, the potential reaction and interaction dynamics are described in more detail, and it indicates where future research could contribute toward addressing this complexity.

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

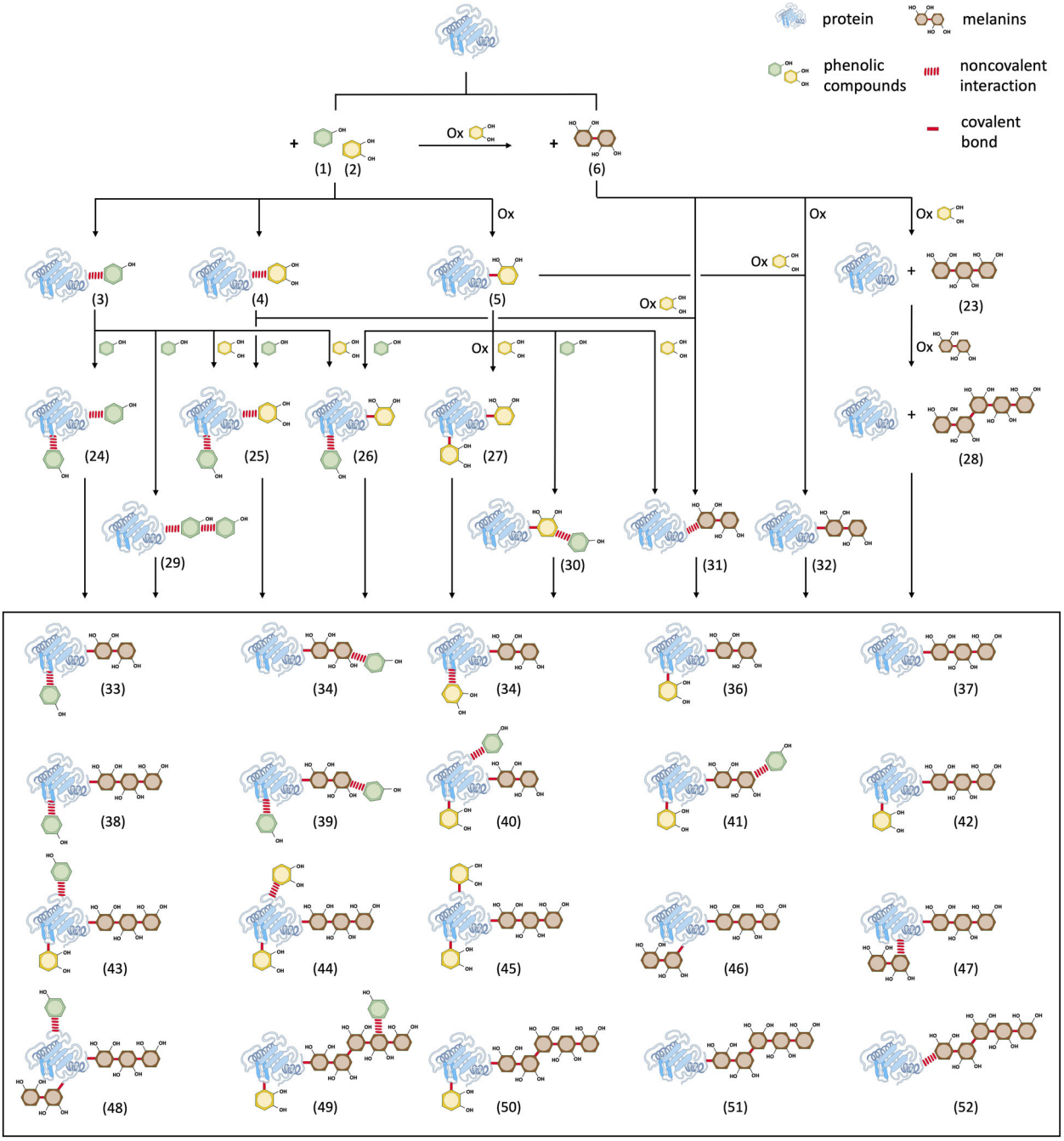

FIGURE 6 Interaction and reaction dynamics of proteins and phenolic compounds. Potential protein structural changes, crosslinks, and evaluations of preferred interaction and reaction pathways are neglected.

After one interaction is accomplished, a new unique scenario for a follow-up interaction/reaction is present. It is a new starting point, where all the consideration given so far must be considered anew. In relatively simple scenarios, as illustrated in Figure 6, this could be either an additional covalent binding to the protein (Figure 6, No. 27) or a subsequent covalent reaction with an already bound PCs, forming a melanin-like compound (Nos. 31 & 32), noncovalent interactions with the protein (Nos. 24–26), or the initially bound PCs (Nos. 29 & 30) as well. All these structures can then be further extended by combinations of covalent or noncovalent binding. Again, noncovalent interactions with a modified protein are possible directly at the protein (e.g., No. 33), at the bound PCs, or at the melanin-like part of the initial modification (e.g., No. 34).

When discussing the presence of proteins and PCs in an oxidative environment, further different scenarios are possible. As already described in Subsection 2.1.2, oxidation might lead either to electrophilic quinones that react preferentially with themselves to form melanins (Figures 1

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

and 6, No. 6) or, to a certain extent, to covalent reaction products with susceptible nucleophilic protein side chains (Figures 1 and 6, No. 5). Between these possibilities, there might also be a certain probability that small melanins (dimers, trimers, etc.) are formed primarily, which can then react with a protein side chain (Nos. 31 & 32). Moreover, melanins can be extended by subsequent addition of a (single) PC (No. 23) or reactions between melanins (No. 28). The latter must not necessarily undergo a covalent reaction with a protein (e.g., Nos. 42, 46, & 51). It seems to be more likely that melanins of increasing molecular size ("polyphenols") are progressively more susceptible to noncovalent protein–phenolic interactions (Nos. 31 & 52). Comparable observations have been made for condensed tannins (Kumar & Horigome, 1986). Theoretically, mixed interactions are conceivable as well (Nos. 38–41 & 47).

In complex systems, the diverse protein–phenolic interaction mechanisms and reaction pathways—especially the complexity of their dynamics—must be considered. The question of the initial reactants' interaction sequence must be addressed: Do PCs interact primarily with proteins or with other PCs to form melanin-like structures initially? Notably, all interactions and reactions mutually influence one another, with each resultant product serving as a precursor for subsequent pathways. Particularly noteworthy is the role of protein unfolding induced by interaction or reaction, which subsequently facilitates further interactions or reactions via various pathways. In Figure 6, the latter aspect was neglected to maintain simplicity. Quantitative analysis of the various products is challenging due to their variable reactivity and potential for immediate further reactions. Further reactions underscore the vast array of interactions and reactions, which are characteristic of prominent food chemical processes such as the MAILLARD reaction (Hellwig & Henle, 2014) or lipid peroxidation (Johnson & Decker, 2015). In fact, the resemblance to the MAILLARD reaction appears to be evident, as all three main stages are present and comparable: (I) an initial phase, followed by (II) subsequent reactions yielding an array of intermediate products. Finally, (III) the formation of covalently bound PCs may also lead to protein crosslinking via distinct phenolic bridges, representing an additional pathway for complex molecular rearrangements (cf. Section 4). The overwhelming diversity of products has severe technological and nutritional consequences.

4 | CROSSLINKING FURTHER COMPLICATES INTERACTION AND REACTION DYNAMICS

While the preceding Section 3 elucidated the dynamics of protein–phenolic interactions, it is also conceivable that proteins may engage in further interactions utilizing intermediate products as a starting point, leading to the formation of protein–phenolic crosslinks. While noncovalent protein–protein interactions adhere to typical protein chemical mechanisms (Cusick et al., 2005; Wodak et al., 2013), the generation of covalent protein–phenolic crosslinks necessitates oxidative conditions once again and is typically catalyzed by the same enzymes or occurs autoxidatively as previously described in Subsection 2.1.2. Figure 7 presents a simplified overview of crosslink formation. In addition to formation via single-subunit phenolic molecules (No. 53), melanins may initially form, subsequently undergoing a protein reaction with ensuing crosslinking. Furthermore, two modified proteins (with at least one PC bound) could interact, resulting in melanin-like bridges (e.g., Nos. 54–56). In such instances, phenolic structures with more than one aromatic ring may furnish the requisite binding sites following prior oxidation to serve as a bridge between two proteins, as depicted as No. 55. Examples of such naturally occurring structures typically arise from the group of flavonoids, including, among others, the tea flavonoid gallates (e.g., epicatechin gallate or epigallocatechin gallate) (Chen et al., 2011; Tan et al., 2022). Similarly, an initial dimerization of a single PC may, in turn, provide sufficient sites for crosslinking, which can induce both noncovalent and covalent interactions (Nos. 54 & 56).

Additionally, further complexities in crosslinking dynamics are inherent within the initial PC structure, particularly in the capacity of certain PCs to adopt isomeric forms. These PCs can subsequently interact with proteins in varying manners, resulting in diverse crosslinking outcomes. This scenario is exemplified by chlorogenic acid, which can exist in different positional isomeric forms (3-, 4-, or 5-chlorogenic acid) in plant-derived material, and these positional isomers can additionally undergo *cis/trans* isomerization, depending on physicochemical conditions. Due to these variations, a multitude of interaction and reaction possibilities can occur between proteins and PCs, thereby complicating the description and monitoring of their crosslinking products within food matrices.

To comprehend the process of crosslinking and elucidate its mode of action, a thorough understanding of how crosslinking progresses, the primary factors influencing crosslinking, and the specific reaction sites and types of crosslinking involved is necessary. Given the anticipated high structural diversity of available PCs, crosslink formations are expected to vary accordingly, yet their full elucidation remains pending. However, the majority of chemical structures of reaction products, particularly those involving exogenous PCs, have not been identified, and those predicted remain analytically unrecorded.

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

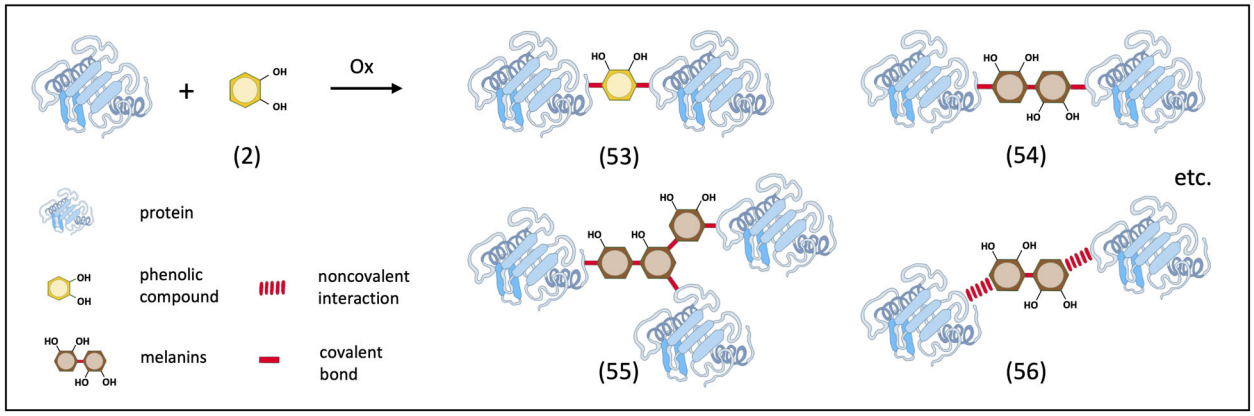

FIGURE 7 Crosslinking interactions and reactions of proteins and phenolic compounds. Potential protein structural changes are neglected for the sake of simplicity.

Consequently, there is a demand for additional complementary strategies, such as labeling of reaction sites and affinity mapping. Furthermore, the accessibility of potential reaction sites can be explored using double-headed agents, while degradation can be employed to simplify subsequent analysis and identification of reacting crosslinked peptides.

Protein crosslinking by PCs is compounded by various other food ingredients, expanding the complexity further (cf. Subsection 2.3 and Section 3). Food proteins embedded within complex matrices undergo diverse chemical modifications, including acetylation, glycosylation, and phosphorylation. These modifications directly impact the structural properties of proteins, altering their interaction characteristics and reaction kinetics with PCs. For instance, reports exist about hetero-crosslinking between proteins and polysaccharides, involving endogenous tyrosine side chains and exogenous PCs (Piber & Koehler, 2005; Selinheimo et al., 2008).

5 | PROTEIN–PHENOLIC INTERACTION AND REACTION PRODUCTS RESULT IN VARIOUS PROPERTIES

So far, the principles of protein–phenolic interactions and reactions, the comprehensive range of interaction and reaction products, and, thus, the inherent complexity in dependence on the prevailing physicochemical conditions have been presented. This section focuses on the consequences, possibilities, and limitations in relevant biological systems, that is, food. More specifically, it describes how the reaction products influence the sensory perception of foods rich in PCs, as well as what is known about the sensory effects of PC–protein modifications (Subsection 5.1), the technological properties of the proteins (Subsection 5.2), and the impact on the digestibility of the food and physiology (Subsection 5.3).

5.1 | Sensorial properties

PCs are well known for their strong effect on sensorial properties in foods. They are linked with bitterness and astringency perception in various foods such as wine, cocoa, and tea (Bittner, 2006), but also with color changes such as the browning of fruits and vegetables (Murata, 2022) and especially the black color of fermented tea (Bittner, 2006). PC interactions with amino acids, peptides, or proteins can affect these properties, but due to the complexity of the reactions, the outcome is still not well understood thus far (cf. Subsections 2.1 and 2.2).

5.1.1 | Bitter and astringent taste

Several PCs like epigallocatechin gallate, coumaric acid, or procyanidins are described as bitter (Ferrer-Gallego et al., 2014; Soares et al., 2018). In contrast, the perception of astringency in various foods, such as wine, cocoa, and tea (Bittner, 2006), seems to be only possible when larger polyphenols interact with proline-rich saliva proteins in the oral cavity (Pires et al., 2020; Rossetti et al., 2008). This is thought to be a direct consequence of the noncovalent interaction of PCs with the proline-rich proteins in saliva and the oral mucosa epithelium (Noble, 2002), primarily mediated by PCs that are not already covalently bound in the food (cf. Subsection 2.2.2). The astringent sensation that PCs imparted to cocoa, for example, may be mitigated during the processing of cocoa beans: Oxidation of PCs could lower the content of sensory-active PCs not only through exudation but also through subsequent protein–phenolic interactions (Misnawi et al., 2005). Similarly, a

1541 4337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

reduction in the bitterness of oleuropein, apigenin, and luteolin, as well as the astringency of oleocanthal, has been observed in olive oil in the presence of proteins, which is attributed to interactions between PCs and proteins (Peyrot des Gachons et al., 2021). However, the extent to which covalent protein–phenolic reaction products differ from noncovalent interactions in terms of flavor impact has not been clearly elucidated and should be investigated in the future.

5.1.2 | Formation of colored compounds

In addition to taste perception, reactions of oxidized PCs with amino acids may lead to novel compounds with a broad array of colors, which are promising candidates for use as food colorants. It is well known that the alkaline extraction of proteins from defatted sunflower meal gives rise to intense greening of the proteins, which is caused by the oxidation of chlorogenic acid and subsequent formation of benzacridines (Ishii et al., 2021). The green color hampers the application of sunflower protein for food use (Wildermuth et al., 2016), especially when light-colored products are to be developed (Lo Verde et al., 2023). Benzacridine formation is initiated by the oxidative dimerization of chlorogenic acid, followed by the addition of the amino group and subsequent cyclization (Yabuta et al., 2001). When the α-amino group reacts with the chlorogenic acid dimers, the benzacridines bear only the amino group, whereas the reaction of, for example, the amino group of β-alanine or the ε-amino group of lysine yields benzacridines with the entire amino acid attached (Bongartz et al., 2016; Drucker et al., 2023). While in most cases green hues caused by benzacridines are developed, oxidative coupling of chlorogenic acid with tryptophan leads to the formation of an intense red color. No color is formed when the thiol group of cysteine is involved in the reaction (Bongartz et al., 2016). The red color is caused by a cyanine-type chromophore of remarkable stability toward pH changes and thermal treatment (Moccia et al., 2021). More recent studies demonstrated the formation of a complex compound bearing one chlorogenic acid and two tryptophan moieties (Santarcangelo et al., 2023). Subsequent investigations into its physicochemical properties and application studies suggest its suitability as a food colorant (Santarcangelo et al., 2024).

The formation of benzacridines is favored at higher pH values, and cysteine has been shown to inhibit green color development (Atonfack et al., 2019; Liang & Were, 2020). The green color turns yellow under reducing conditions, for example, in the presence of ascorbic acid (Yabuta et al., 2001). As greening has also been prevented with sulfites (Wildermuth et al., 2016), any recipes containing this additive are challenging when the formation of benzacridines is desired. Enzymatic hydrolysis of chlorogenic acid has been demonstrated to be another technological strategy to avoid green color formation (Lo Verde et al., 2023).

Most studies published so far deal with the reactions of free amino acids, which yield the abovementioned reaction products. However, investigations modifying β-lactoglobulin with chlorogenic acid under alkaline conditions revealed that benzacridines also occur in protein-bound form (Ali et al., 2013). The results obtained from the alkaline treatment of sunflower meal used as ruminant feed also indicate the presence of protein-bound benzacridines (Bongartz et al., 2018). Obviously, the ε-amino group of lysine represents the preferred sites of derivatization. Given that *N*-terminal amino acids, unless derivatized, by definition bear a free amino group, it is reasonable to assume that analogous reactions may happen with peptides and proteins in general. Although preliminary in vitro assays indicate a lack of toxicity of free benzacridines (Iacomino et al., 2017), studies on the metabolic fate and toxicological properties of bound benzacridines have not yet been conducted.

The studies conducted so far on benzacridines and other colored compounds formed by oxidative coupling of chlorogenic acids and amino acids suggest promising properties of these compounds for potential use as food colorants. There is a lack of knowledge about the presence of protein-bound colored reaction products—in particular, their binding sites and the extent of protein modification. It is even conceivable that benzacridines mediate the crosslinking of proteins. The conditions required for these modifications to form have yet to be determined. Proteins containing benzacridine moieties can either be used directly as colorants or serve as substrates for enzymatic proteolysis. This process generates peptides with unique technological, even also biological properties that may extend beyond just providing color. In addition to comprehensive technological studies that need to be performed to demonstrate the potential of the pigments as food colorants, their legal status needs to be addressed.

5.2 | Changes in technological properties of proteins

PCs may affect the protein's technological properties, that is, foam stabilization, emulsion stabilization, and gelation. The performance of proteins in the formation of these (food) systems is dictated by the proteins' solubilization, structure, surface hydrophobicity, charge, and size. Although similar to the previous findings on the effect of PCs on protein structure (cf. Subsections 2.1 and 2.2;

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

Figure 4), there are also inconsistent results about the impact on the technological properties.

5.2.1 | Foams and emulsions

Foams and emulsions play a crucial role in providing structures in food. In such multiphase systems, two immiscible phases, gas–water or oil–water interfaces, are present. Proteins possess a high capacity to stabilize such systems, as their amphiphilic properties (presence of hydrophobic and hydrophilic amino acids) enable them to stabilize the air–water or oil–water interface by adsorption, followed by intra- and intermolecular interaction among adsorbed proteins (Kieserling et al., 2021; Narsimhan & Xiang, 2018). To achieve proper interface stabilization, proteins usually need to have high solubility, structural flexibility, and preferably a non-aggregated (or native) state in the quaternary structure (Sagis & Yang, 2022). Small and soluble proteins can quickly adsorb at the interface, giving small air bubbles and oil droplets. Then, protein–protein interaction is necessary to form a viscoelastic interfacial protein film, which results in high stability against foam collapse and oil droplet coalescence (Foegeding et al., 2006; Kieserling et al., 2021). There is a strong indication that hydrophobic interactions among proteins play a major role in protein–protein interactions at the interface. Other possible key properties are protein flexibility to structurally orient interaction sites toward each other and the formation of a dense protein film, which can, for instance, be achieved with low repulsive (electrostatic) forces between adsorbed proteins (Cao et al., 2018; Kieserling et al., 2021).

Protein–phenolic interaction can strongly affect surface activity, depending on the type of protein, PC, mode of binding (covalent vs. noncovalent), level of modification, and crosslinking. Thus, it is extremely difficult to extrapolate common effects.

For noncovalent interactions, there are reports about increased foam ability and stability of whey protein isolate, after adding gallic acid and epigallocatechin gallate (Cao et al., 2018) or chlorogenic acid (Jiang et al., 2018). In both cases, this was explained by a decreased surface hydrophobicity and increased solubility of the protein. For sunflower proteins, the noncovalent interaction with chlorogenic acid resulted in the crosslinking of 4- to 100-nm-large aggregates (Karefyllakis et al., 2017). While the surface tension of the mixture was lower than that of the protein alone, the oil droplet size was larger, which could indicate droplet flocculation.

Reports also show that unbound PCs can compete with proteins for the interface, weakening the interfacial film and destabilizing the foam stability. For example, Yang et al. (2021) found that sinapic acid–whey protein isolate mixtures lowered interfacial stiffness and foam stability, hindering protein–protein interaction at the interface. Similar findings were reported for rapeseed protein-stabilized emulsion droplets. Free sinapic acid interrupted the interfacial film, and the crosslinking of proteins induced by sinapic acid also resulted in overall weaker interfacial films and enhanced coalescence (Ntone et al., 2022). Bock et al. (2022) tested different phenolic acid derivatives for their effect on protein-stabilized interfaces and found that they can result in weaker interfacial films (O/W) and more flocculation of droplets. In contrast, PCs can also impact larger aggregated structures, such as protein amyloid-like fibrils. Yan et al. (2024) demonstrated that the emulsifying properties of amyloid-like fibrils of lentil proteins were enhanced by noncovalent interactions with epigallocatechin gallate. The PCs altered the shape of the fibrils from an elongated to a more aggregated structure, resulting in stronger interactions at the oil–water interface, likely due to more exposed hydrophobic groups, ultimately leading to increased emulsion stability. This enhancement was attributed to a more balanced hydrophobic/hydrophilic ratio of the products (Yan et al., 2024). Thus, for noncovalent interactions, both positive and negative effects were observed. Here, positive effects were usually explained by increased solubility of the modified proteins, while negative effects were usually caused by unbound PCs and crosslinking effects, which could potentially be controlled by controlling the protein:PC ratio.

However, crosslinking does not always result in decreased interface stability. For example, the foam stability was improved for lactoferrin–tannic acid mixtures that formed aggregates of 30–70 nm. It was assumed that this was due to particle pinning and thin film stabilization. Also, von Staszewski et al. (2014) found that aggregates between β-lactoglobulin and green tea PCs gave higher emulsion stability than pure β-lactoglobulin, explained by Pickering-like stabilization or the formation of a thick interfacial film. Thus, further research is required to elucidate the effect of PCs on the aggregation of proteins and the resulting interfacial stabilization mechanisms.

With regard to covalent modifications, the results are less divergent than for noncovalent modifications, as mentioned previously, due to the higher stability of the interaction, making it more controllable. The covalent modification of proteins affected the solubility and the flexibility of the protein, which increased the overall emulsifying properties of plant proteins, with some discrepancies caused by using different conditions such as preheating. The most relevant studies are summarized by Keppler et al. (2024).

Compared to single-phase water systems, the primary challenge in comprehending the influence of PCs on

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

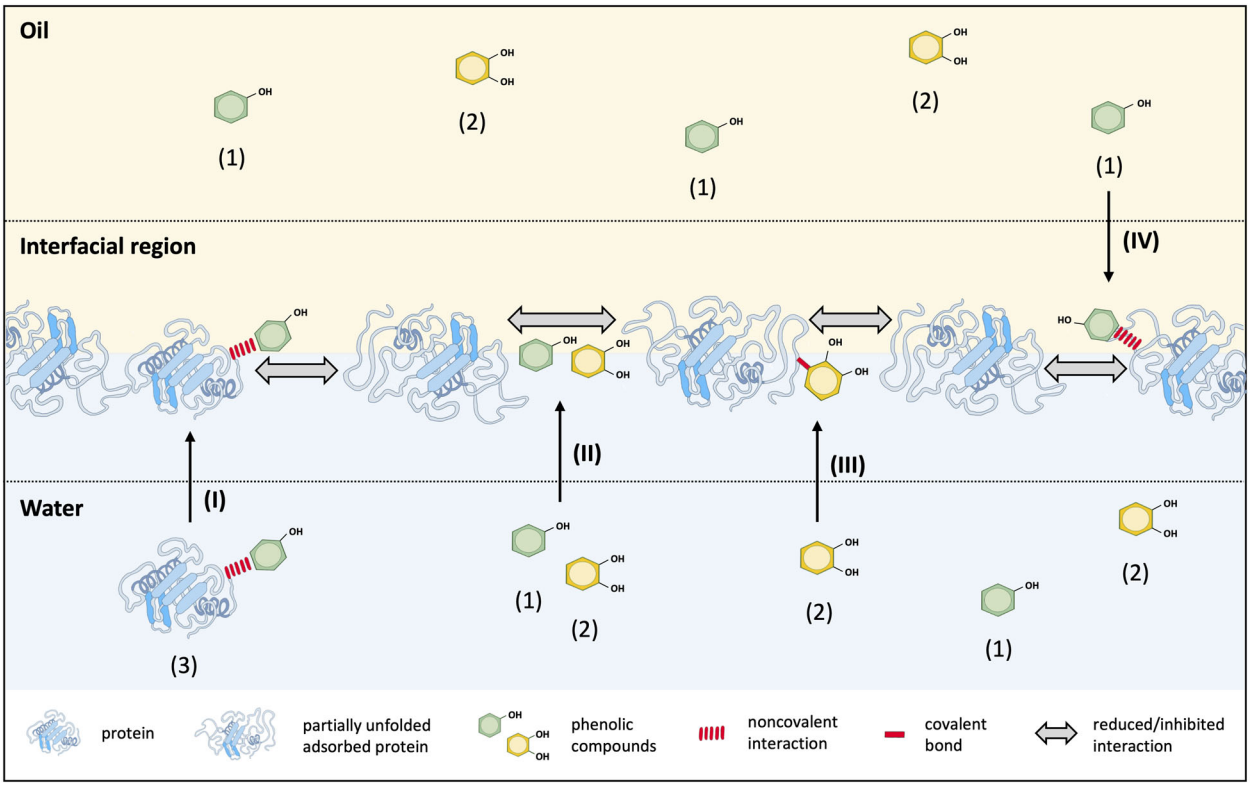

FIGURE 8 Potential locations of phenolic compounds (PCs) during the interfacial stabilization process. The PCs may exist (I) in the bulk water phase, then interact or react with the protein prior adsorption, (II) at the interface in a nonbound free form or may interact/react with adsorbed proteins (III) from the bulk water phase or (IV) from the oil phase, following Bock et al. (2024).

protein-stabilized interfacial systems lies in the increased complexity resulting from the presence of an additional oil phase, thereby leading to a multitude of interaction sites. As illustrated in Figure 8 for an oil–water interface, the PC can (I) interact with the protein within the bulk water phase and then adsorb at the interface in a protein-bound state, (II) adsorb at the interface in a nonbound free form (i.e., especially amphiphilic PCs with polar and unipolar moieties), or interact with the already adsorbed protein from (III) the bulk water phase or (IV) the oil phase.

For example, the interaction and reaction of proteins with PCs in the bulk water phase (I) before adsorption might influence the protein structure (e.g., hydrophobicity) and subsequently its interfacial behavior. Therefore, systematic studies of the interfacial behavior should include structural analyses of the interaction and reaction products, enabling conclusions regarding their influence on emulsion and foam stability. However, certain (plant) proteins exhibit complex structural characteristics even prior to undergoing interactions and reactions with PCs. For instance, the solubility of plant proteins is frequently quite low even before engaging with PCs and may further diminish due to protein–phenolic interactions or reactions. This presents a significant challenge for interfacial studies focusing on such interactions or reactions, as analysis of the resulting insoluble products is often unfeasible using conventional methods.

In terms of the interface, all remaining interaction sites of the PC (II–V) possess the potential to inhibit the formation or to disrupt the already existing interfacial protein–protein interactions, leading to weak interfacial protein films and, consequently, low stability of oil (or air) droplets, as exemplified by Bock et al. (2022) through the enhancement of droplet flocculation of emulsions. Ultimately, the distribution pattern of PCs underscores the complexity of analyzing its role in interfacial systems, necessitating consideration of its location across all scenarios. The role of unbound PCs after covalent modifications is generally not studied and should definitely be included as a control.

5.2.2 | Gels

Protein gels are three-dimensional network structures entrapping a liquid, which are supported by intermolecular covalent and noncovalent protein bonds. There are different types of gels, but the most common ones in food are those induced through heating, subsequent protein denaturation (i.e., structure unfolding), and crosslinking

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

(Nicolai & Chassenieux, 2019). The formation of heat-induced gels occurs in three steps: (I) unfolding and dissociation of proteins, (II) aggregation of the unfolded proteins due to interactions between newly exposed groups (e.g., hydrophobic or disulfide groups), and (III) the interaction of aggregates to form a space-filling gel network. To form stiff gels, the proteins should be well-distributed in solution, which requires highly soluble proteins with a repulsive electrostatic charge (Chen et al., 2016). Also, the aggregated state plays a key role, as native and non-aggregated proteins are most suitable to induce the two-step aggregation to form the homogeneous gel network. Extensive pre-aggregation of proteins might give more heterogeneous gels, which could be weaker and more brittle, also due to fewer interaction sites per unit of volume compared to smaller and more native proteins.

For noncovalent modification, crosslinking was found to be especially detrimental to a homogeneous gel structure. Chen et al. (2021) investigated pea protein mixtures with green tea PCs and observed increasing concentrations of inhomogeneous clusters with increasing additions of green tea PCs. This resulted in a poorly distributed network structure and, thus, weaker gels. They postulated that the presence of PCs disrupts a continuous protein–protein gel structure. Also, Jia et al. (2022) found that the addition of chlorogenic acid to sunflower protein had a negative effect on the gel strength, and similar findings were observed for canola protein gels after the addition of sinapic acid or thomasidioic acid (Rubino et al., 1996).

However, when covalently modifying sunflower protein with chlorogenic acid, a slight increase in gelling properties was observed, which then declined sharply when the protein was modified more strongly (Jia et al., 2022). A strong level of covalent modification likely blocks all cysteine residues with PCs, thereby disrupting network formation (Keppler et al., 2020). While there are not yet many studies on the effect of PCs on the gelling properties of proteins in food, there are reports about utilizing the interaction for making bioadhesives and glues with additional antioxidative or antibacterial properties. In these reports, the bioadhesives are called "bioinspired" or "mussel-inspired," because the wet adhesion properties of mussel foot proteins are caused by tyrosine oxidation to L-DOPA, with catechol groups found to be the cause of their strong glue effect. The addition of PCs, such as caffeic acid, chlorogenic acid, or gallic acid, to denatured soy proteins at alkaline pH was postulated to mimic these effects after forming covalent interactions between the protein and the PCs. While for chlorogenic acid and caffeic acid, the viscoelastic properties of the soy protein isolate gel decreased, they increased for gallic acid. Thus, different PCs had different effects on the protein gels, probably due to the level of modification and type of interaction. When adding PCs to preformed soy protein–agarose hydrogels, the PCs acted as plasticizers, resulting in stretchable material with interesting mechanical properties (Argenziano et al., 2023). There are also other applications, where the addition of PCs can be used to modulate protein network properties. For example, polyphenols were found to interact with functional amyloid protein aggregates from lysozyme, soy, or whey, inducing the formation of supramolecular structures that can be used as (food) hydrogels of high mechanical stability (Hu et al., 2018; Nian et al., 2022; Xu et al., 2022).

There is limited research available concerning the gelation of proteins in the presence of PCs. The presented information is largely generalized, with some discrepancies stemming from methodological variations, as well as differences in PCs and protein types. Nonetheless, it is noteworthy that noncovalent interactions between proteins and PCs appear to hinder gel network formation, as evidenced by the formation of heterogeneous clusters at higher interaction levels, while covalent interactions at lower levels tend to give stiff gels.

A significant challenge arises in experimental setups where protein modification levels are increased, necessitating a substantial excess of PCs to protein. Even with extensive filtration and dialysis aimed at excluding noncovalent modifications, complete removal of all noncovalently bound PCs from covalently modified proteins remains elusive. Consequently, these residual PC molecules often contribute to observed gelation effects, complicating interpretation.

Beyond these experimental challenges, gel formation is inherently complex, with various gel networks arising from different forces, such as prevalent covalently linked gels versus those primarily stabilized by hydrophobic interactions. The addition of PCs can thus yield diverse effects, contingent upon the specific proteins, PC types, protein:PC ratios, and the underlying network structure of the gel.

5.3 | Changes in physiological properties

While Subsections 5.1 and 5.2 addressed the impacts of protein–phenolic interactions and reactions on food properties prior to consumption, the following subsections provide an overview of their effects postconsumption. Digestibility and bioavailability of proteins and PCs are the main determinants for their bioactivity, where interactions or reactions may potentially interfere (cf. Subsection 5.3.1). Another relatively new physiological consideration is the influence of PCs on protein allergenicity (cf. Subsection 5.3.2).

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

5.3.1 | Digestibility and bioavailability of protein interaction and reaction products

Many studies showed that the addition of PCs to proteins alters protein digestibility. In the case of noncovalent bonds, digestibility can be affected by interactions of PCs with dietary proteins or with proteases of the gastrointestinal tract (Ertop & Bektaş, 2018). In fact, interaction with PCs can either stabilize or destabilize (i.e., induce partial unfolding) the protein structure (Kanakis et al., 2011) or sterically hinder the access of proteases so that protein digestibility is reduced. A partial unfolding of the protein structure would in turn increase protein digestibility (Hasni et al., 2011).

Several studies investigated the effects of protein–phenolic interactions on in vitro digestion of isolated proteins by specific proteases. Regarding pepsin, the digestibility of β-lactoglobulin was decreased in the presence of PCs from green and black tea, coffee, or cacao (Stojadinovic et al., 2013). In contrast, Tantoush et al. (2012) found an increasing effect of green tea PCs on the digestibility of different proteins such as β-lactoglobulin or peanut proteins. The peptic digestibility of soy protein and egg white gradually increased with increased amounts of grape seed PCs (Yu et al., 2016), whereas for casein, there was an increase at low concentrations and a decrease at higher concentrations of PCs. Digestibility with trypsin was reduced for casein and soy protein isolate by grape seed PCs (Yu et al., 2016). The same authors found reduced digestibility of proteins from bread with grape pomace PCs added, after using pepsin followed by trypsin. Furthermore, for tannins with a molecular weight of over 500 Da and condensed tannins, a negative effect on the digestibility of bovine serum albumin has been reported (Kumar & Horigome, 1986).

Regarding the effect of PC interactions on the activity of the gastrointestinal proteases, the proteases pepsin, trypsin, chymotrypsin, and elastase have been studied most frequently. The interaction of PCs with proteases is conceivable in various ways: in principle, binding of PCs can lead to a partial unfolding (in the case of bulky PCs) or stabilization of the enzyme structure and, thus, changes in their activity. On the one hand, this can be accompanied by a reduction in enzyme activity, and, on the other hand, proteins that serve as substrates could better fit the catalytic center. Furthermore, competitive inhibition can occur at the catalytic or allosteric center (Cirkovic Velickovic & Stanic-Vucinic, 2018). Some studies showed significant inhibition of pepsin by various PCs (e.g., epigallocatechin gallate, catechin, and tea extracts) and different protein substrates. Their effect on pepsin was different in the various studies; both inactivation of the protease and improved digestion of β-lactoglobulin were observed (He et al., 2007; Tagliazucchi et al., 2005). Different results were also shown for chlorogenic acid in the digestion of lysozyme and soy protein isolate by pepsin and for coffee extract in the degradation of β-lactoglobulin (Rawel et al., 2000, 2002). In contrast, in these studies, polymeric tea PCs and hydroxycinnamic acids have a consistently inhibitory effect on trypsin, chymotrypsin, and elastase in these studies. The trypsin activity was significantly decreased by tea PCs, but not completely inhibited (Huang et al., 2004).

The covalent modification of proteins with various PCs (Kroll et al., 2000; Rawel et al., 2000) resulted in a higher resistance of the proteins toward digestion by gastrointestinal proteases (Rawel, H. M., Kroll, J., & Hohl, U. C., 2001). Above all, covalent bonds occur for the ε-amino group of lysine or thiol-group of cysteine (cf. Subsection 2.1.2). Thus, binding to ε-amino groups of lysine residues could prevent the binding of trypsin, resulting in digested peptides with a high molecular weight (Zee & Garcia, 2012). However, at higher levels of chlorogenic acid attached to bovine serum albumin, Rawel et al. (2002) demonstrated that tryptic digestion is increased compared to low levels. This might be due to high levels of chlorogenic acid partially unfolding the protein structure, thereby facilitating the tryptic attack. An in vivo study of soy protein showed that the *protein digestibility corrected amino acid score* (PDCAAS) values dropped stronger for lysine, tryptophan, and sulfur-containing amino acids at high versus low derivatization levels with chlorogenic acid (Rohn et al., 2006). Thus, protein–phenolic interactions undoubtedly affect protein digestibility, although reports on the exact nature and extent of these effects remain discrepant.

In numerous studies, precise differentiation between the effects of PCs on dietary protein and protease activity remains challenging. Typically, only the final concentration of soluble peptides (e.g., solubility in trichloroacetic acid) is evaluated, mainly neglecting critical parameters such as peptide length or their identification. Ongoing developments in untargeted peptide analysis using mass spectrometry and automated annotation will help to provide more detailed data in the future (Vreeke et al., 2022). Moreover, studies often display inconsistencies due to varied experimental conditions and inadequate exploration of factors such as the initial and modified protein/protease structure, direct interactions with PCs, or the extent of modification. Figure 9 illustrates the impact of PC-induced interactions and reactions on peptide composition, directly affecting peptidase binding sites (Figure 9, No. 58) or indirectly through protein structural changes exposing additional binding sites (No. 59), compared to unbound proteins (No. 57). Consequently, protein–phenolic interactions may either impair (No. 61) or enhance (No. 62) the protein digestibility. Furthermore, many studies focus on a

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

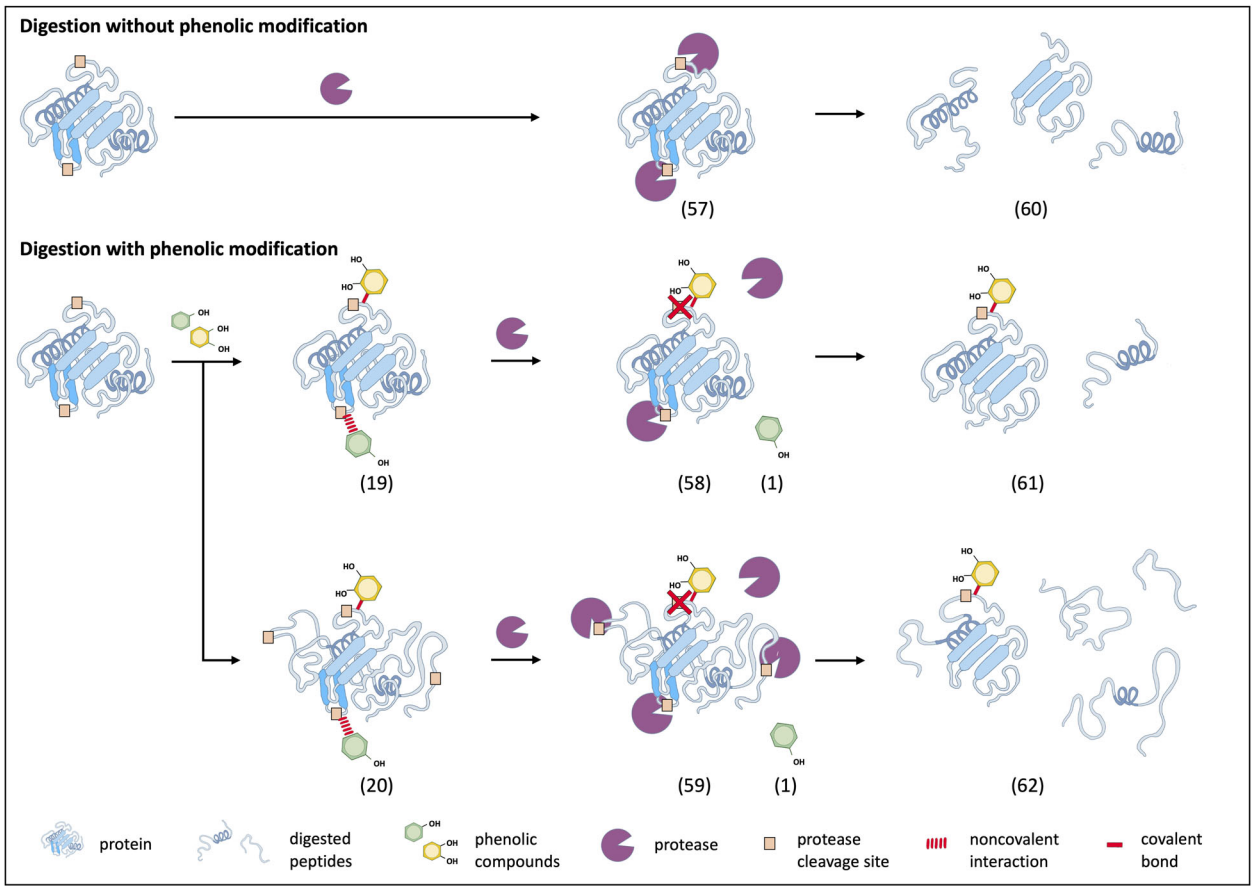

FIGURE 9 Overview of different possibilities regarding the enzymatic digestion of proteins with and without interaction or reaction with phenolic compounds as well as the respective formation of different peptide schemes.

single protease, providing mechanistic insights but failing to replace in vivo tests that are crucial for determining the *digestible indispensable amino acid score* (DIAAS) or the PDCAAS. The in vivo digestive environment encompasses diverse conditions (e.g., ionic strength, pH) and competing interactions and reactions, often overlooked in laboratory settings. For instance, salivary proteins may compete with dietary proteins and digestive enzymes, leading to the formation of protein aggregates within the digestive tract (Morzel et al., 2022).

5.3.2 | Phenolic compounds may affect protein allergenicity

When PCs interact with protein epitopes, the interactions, especially covalent reactions, may influence the proteins' allergenicity and hence food allergy (Pan et al., 2022; Singh, 2011). Protein epitopes comprise either a specific sequence of amino acids within the primary structure ("sequential epitopes") or a three-dimensional arrangement of amino acids, that is, secondary and tertiary structure ("conformational epitopes"). Consequently, protein allergenicity may be affected both directly by protein–phenolic interactions and reactions and indirectly by changes in the proteins' structure and, thus, the linear and the three-dimensional epitopes.

Pessato et al. (2018) described a significant reduction in the binding of monoclonal immunoglobulin E antibodies to whey protein after noncovalent interaction with epigallocatechin gallate. The authors explained the reduction of phenol interactions with a single amino acid within the whey proteins' primary structure, that is, proline and the subsequent structural changes. Regarding the secondary structure, Pu et al. (2021) described an α-helix-to β-sheet transition of β-lactoglobulin after interaction with different PCs. Hereby, noncovalent interactions with phloretin showed the highest reduction in α-helix from 49.5% to 8.2%, thereby clearly decreasing the allergenicity. Liu et al. (2021) used the flavonoids luteolin, myricetin, and hyperoside and suggested covalent reactions with β-lactoglobulin. While the molecular mass of the reaction products increased, the immunoglobulin G or immunoglobulin E binding activity decreased. The experiments on rat mast cells of the RBL-2H3 line showed a decrease in β-hexosaminidase and histamine release,

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

indicating a reduction in the allergenic effect of the protein–phenolic reaction products described. Furthermore, the secondary structure of the proteins was partly unfolded compared to β-lactoglobulin without phenol interactions. Thus, the authors assumed that β-lactoglobulin–flavonoid reaction products influence T-cell activity and, therefore, attenuate the allergic response. Another way to affect protein epitopes is to form insoluble reaction products between the epitopes of gliadins (the proteins that cause wheat allergy) by adding phenolic-enriched extracts of apples, cranberry, and artichoke (Pérot et al., 2017). The authors explained the lowered allergenic effect by a potential formation of covalent protein–phenolic reaction products, thereby affecting the proteins' epitopes. However, masking by noncovalent interactions cannot be excluded.

Some observations suggest that protein structural changes induced by PCs may not necessarily correlate with decreased protein allergenicity. Indeed, certain studies have proposed that structural modifications of epitopes could potentially enhance the allergenicity of proteins. For instance, Davis and Williams (1998) demonstrated that basic physical treatments, such as heating and the consequent unfolding of β-lactoglobulin, led to the exposure of previously concealed epitopes. Additionally, chemical modifications like β-lactoglobulin glycation enhanced the affinity of immunoglobulin E antibodies compared to unmodified β-lactoglobulin (Corzo-Martínez et al., 2010). Furthermore, allergic reactions involve intricate cascades typically triggered by entire protein groups (e.g., milk allergy involves the milk protein group of caseins and all whey proteins), whereas PCs tend to mask only individual epitopes of specific proteins. Thus, to mitigate allergic reaction cascades, such as milk allergy, a comprehensive understanding of the specific interactions and reactions between PCs and epitopes, along with resultant structural alterations of the protein, is essential. However, achieving such depth of understanding is currently challenging. For instance, the reliable and reproducible occurrence of structural changes in sequential and conformational epitopes remains elusive, as previously discussed in several sections. Moreover, beyond the realm of chemical mechanisms, it is necessary to devise suitable methodologies to ascertain whether the modified epitopes retain recognition beyond enzyme-linked immunosorbent assay, such as through evaluation with human blood sera, which often poses an obstacle. Additionally, determining the degree of modification and stability of resultant reaction products during digestion presents substantial inquiries.

Notably, despite the considerable potential for allergy reduction, comprehensive investigations systematically characterizing the influence of different structural features of PCs on the protein structure and associated modifications of various sequential and conformational epitopes, and thereby protein allergenicity in phenol-rich foods, are prominently absent.

6 | GAINING SYSTEMATIC INSIGHT INTO THE MECHANISMS AND EFFECTS OF PROTEIN–PHENOLIC INTERACTIONS AND REACTIONS OFFERS VALUABLE OPPORTUNITIES

Most interactions and reactions occurring between proteins and PCs happen accidentally during food processing and preparation, depending on the PC and protein composition and the physicochemical conditions. However, with an increasing understanding of the underlying reaction mechanisms, one may even think of a targeted use by directing the influencing factors to enhance or diminish the interactions.

The application and utilization of protein–phenolic interactions in food preparation have received more and more attention in the last few years, whereby the following topics can be observed quite frequently: utilization of both noncovalent interactions and covalently induced protein crosslinks in stabilizing food systems, including emulsions (Zhang et al., 2021) and bakery products properties (Selinheimo et al., 2008). These interactions can be influenced by manipulating the conditions used during food processing and preparation, such as concentrations, pH, and heating (Jia et al., 2022; Zhang et al., 2021), as well as by applying specific (combinations of) enzymes (Isaschar-Ovdat & Fishman, 2018). The attractiveness of applying this concept arises because it could provide enhanced antioxidant effects, higher nutritional values, and more stable products (Zhang et al., 2021). In general, both types of interactions may work synergistically to improve food quality (de Oliveira et al., 2016; Prigent et al., 2003).

However, protein–phenolic interactions may also negatively affect food properties. For example, covalent bonds can lower the nutritional quality of the proteins, especially when essential amino acids are involved (Petzke et al., 2005; Rohn et al., 2006). Moreover, unwanted processes like browning or precipitation can be induced due to the oxidation of PCs and their interactions with proteins. It should be further investigated which degree of protein modification by PCs (i.e., the number of bound PCs per protein) induces meaningful changes in protein properties. Therefore, it is essential to gain further insights into controlling and evaluating such interactions (spontaneous noncovalent interaction and covalent reaction) in dispersed food systems. In studying these interactions, the factors affecting the types and possible combinations of interactions should also be considered in order to

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

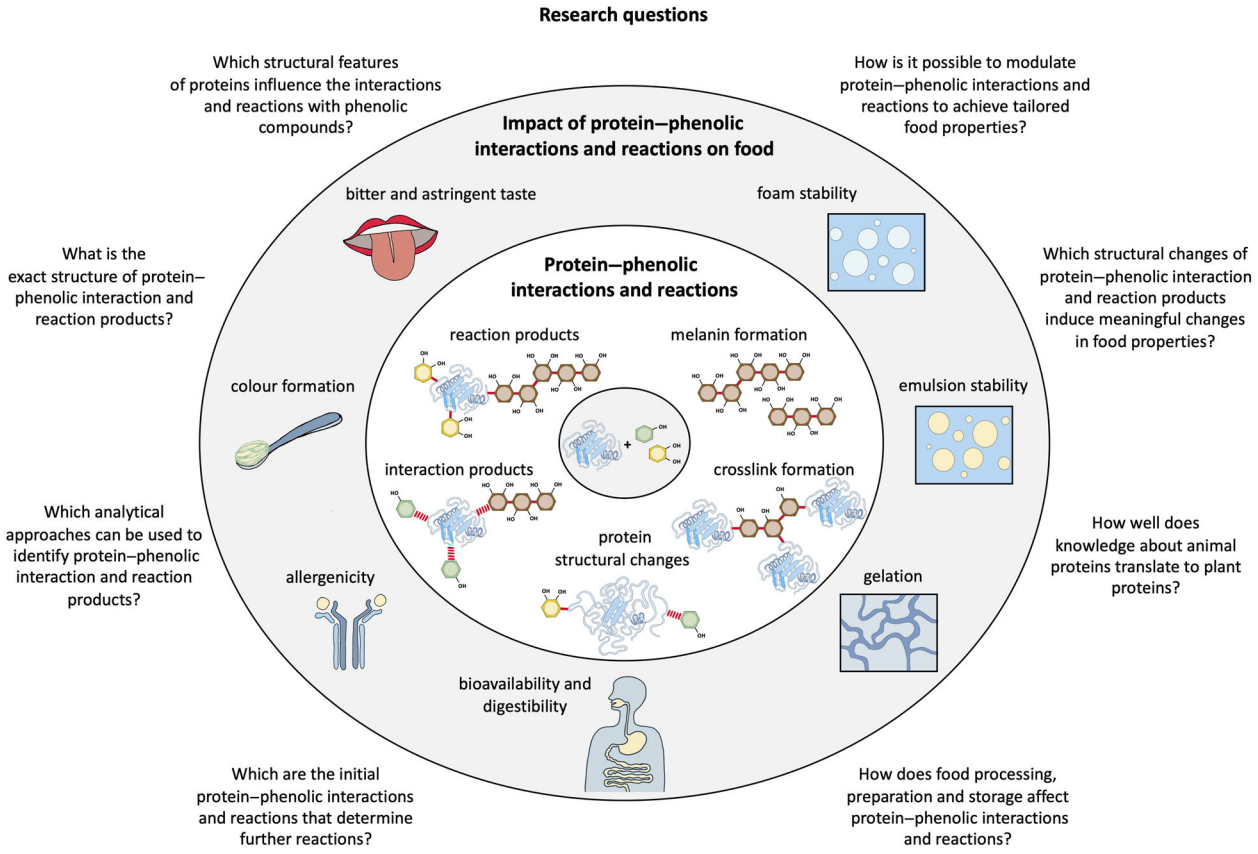

FIGURE 10 Impact of protein–phenolic interactions and reactions on food and potential future research questions.

utilize their respective advantages and limit undesirable effects on protein and phenolic properties (Zhang et al., 2021). Besides this, one should consider, when intentionally adding PCs to food to induce interactions or reactions with proteins, that excessive consumption of PCs (and their quinones) has been associated with adverse health effects (Blaut et al., 2006; Bolton et al., 2000; Cladis et al., 2021).

Further developments are still needed in analytical approaches that can be used as methods to characterize covalent and noncovalent interactions separately and in combination. While trying to gain more systematic insight into the mechanisms involved, the noncovalent interactions are "partly removable," as these are reversible (Prigent et al., 2003; Rawel et al., 2005; Zhang et al., 2021). Moreover, in silico studies serve as valuable complementary methods for characterizing noncovalent interactions, elucidating the mechanisms underlying distinctions such as reversible versus irreversible interactions, competitive versus noncompetitive interactions, and site-specific versus nonspecific interactions (Shahidi & Dissanayaka, 2023). Following covalent reactions is challenging, as the methods available for the characterization mostly rely on breaking down substrates to the level of amino residues or using a multi-enzymatic digestion strategy integrated into a proteomics-based workflow. Both approaches have their specific setbacks. This highlights the need to develop robust analytical approaches to analyze both noncovalent interactions and covalent protein–phenolic reactions.

Currently, it remains challenging to correlate the induced covalent reactions and structural changes in modified proteins or peptides to protein technological and biological functionality. This implies the need for some means of controlling the interactions, predicting possibilities, and tailoring reactions via systematic research. For some PCs, information is now available related to their reactivity toward covalent reactions with different amino acids and peptides (Bongartz et al., 2016; Iacomino et al., 2017). The field of covalent reaction of PCs to proteins still contains many unknowns and should be subject to future scientific studies. More insight is indeed needed to scrutinize the oxidation behavior of the individual PCs, establishing relationships between PC structures and their reactivity toward relevant groups in proteins and identifying their intermediary reaction products, before addressing the final complex mixtures of products resulting from their interactions with proteins. When approaching the situation from the protein side, protein structure and the exposure of the reactive amino acid side chains seem to be the most prominent factors to be addressed.

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

In conclusion, the complexity of the structural and functional changes in protein and PCs during food processing and preparation opens the door to much application potential and opportunities for further work. Especially here, an upcoming research field of interactions of flavor components originating from the complex redox reactions of PCs and their integration into proteins could provide an excellent novel research opportunity (Keppler et al., 2020; Zhang et al., 2021). Moreover, complementary data on the toxicological potential, absorption behavior, further metabolism after intake, and physiological impact of the reaction products will be necessary to allow such protein modifications to become an attractive option and facilitate their more widespread use. Progress toward answering the significant outstanding research questions in this field (Figure 10) will certainly contribute to a better understanding of not only the mechanisms involved in protein–phenolic interactions but will also shed light on how to control such interactions.

7 | CONCLUSION

Beyond traditional literature reviews, this review offers a comprehensive discussion of the current debatable and convoluted facets of the topic of protein–phenolic noncovalent interactions and covalent reactions. A fundamental understanding of the initial interaction and reaction mechanisms concerning the influence of physicochemical conditions and structural features of PCs and proteins exists. Nonetheless, it is evident that a significant knowledge deficit persists concerning subsequent mechanisms regarding interaction and reaction dynamics, the formation of crosslinks, and their consequences for the properties of proteins and PCs. Conversely, understanding of the resulting properties of foods has advanced, thanks to extensive investigations into their sensory attributes, technological properties, and physiological impacts. However, often these effects elude explanation by specific interaction and reaction products due to the considerable structural diversity in product mixtures, which are challenging to analyze. Additionally, it is crucial to recognize the multitude of interactions, reaction pathways, and dynamics, not only between proteins and PCs but also involving other food constituents. These interactions and reactions further contribute to the convoluted dynamics and complex mixture of resultant products. Therefore, comprehensive research in this field requires the consideration of all of these factors to fully understand the\ underlying mechanisms and consequences.

AUTHOR CONTRIBUTIONS

Helena Kieserling: Conceptualization; writing—original draft; writing—review and editing; visualization; validation; project administration. **Wouter J. C. de Bruijn**: Writing—original draft; visualization; writing—review and editing; conceptualization. **Julia Keppler**: Conceptualization; writing—original draft; writing—review and editing; supervision. **Jack Yang**: Writing—original draft; writing—review and editing. **Sorel Tchewonpi Sagu**: Writing—review and editing; writing—original draft. **Daniel Güterbock**: Writing—original draft; writing—review and editing. **Harshadrai Rawel**: Writing—original draft; writing—review and editing; conceptualization; supervision. **Karin Schwarz**: Conceptualization; writing—original draft; writing—review and editing; supervision. **Jean-Paul Vincken**: Supervision; conceptualization; writing—original draft; writing—review and editing. **Andreas Schieber**: Conceptualization; writing—original draft; writing—review and editing; supervision. **Sascha Rohn**: Conceptualization; writing—original draft; writing—review and editing; project administration; supervision.

ACKNOWLEDGMENTS

Open access funding enabled and organized by Projekt DEAL.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

ORCID

Helena Kieserling https://orcid.org/0000-0002-8093-6794
Jack Yang https://orcid.org/0000-0001-8217-1759
Harshadrai Rawel https://orcid.org/0000-0003-2768-1531

REFERENCES

Ali, M., Homann, T., Khalil, M., Kruse, H. P., & Rawel, H. (2013). Milk whey protein modification by coffee-specific phenolics: Effect on structural and functional properties. *Journal of Agricultural and Food Chemistry*, *61*(28), 6911–6920. https://doi.org/10.1021/jf402221m

Argenziano, R., Viggiano, S., Esposito, R., Schibeci, M., Gaglione, R., Castaldo, R., Fusaro, L., Boccafoschi, F., Arciello, A., Della Greca, M., Gentile, G., Cerruti, P., D'Errico, G., Panzella, L., & Napolitano, A. (2023). All natural mussel-inspired bioadhesives from soy proteins and plant derived polyphenols with marked water-resistance and favourable antibacterial profile for wound treatment applications. *Journal of Colloid and Interface Science*, *652*, 1308–1324. https://doi.org/10.1016/j.jcis.2023.08.170

Atonfack, J. T., Ataman, Z. A., & Were, L. M. (2019). Acidulant effect on greening, reducing capacity, and tryptophan fluorescence of sunflower butter cookie dough during refrigerated storage. *Journal of the Science of Food and Agriculture*, *99*(5), 2186–2193. https://doi.org/10.1002/jsfa.9412

Bacon, J. R., & Rhodes, M. J. C. (1998). Development of a competition assay for the evaluation of the binding of human parotid salivary proteins to dietary complex phenols and tannins using

15414337, 2024, 5, Downloaded from https://ifst.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

a peroxidase-labeled tannin. *Journal of Agricultural and Food Chemistry*, *46*(12), 5083–5088. https://doi.org/10.1021/jf980791c

Baxter, N. J., Lilley, T. H., Haslam, E., & Williamson, M. P. (1997). Multiple interactions between polyphenols and a salivary proline-rich protein repeat result in complexation and precipitation. *Biochemistry*, *36*(18), 5566–5577. https://doi.org/10.1021/bi9700328

Bijlsma, J., de Bruijn, W. J. C., Hageman, J. A., Goos, P., Velikov, K. P., & Vincken, J.-P. (2020). Revealing the main factors and two-way interactions contributing to food discolouration caused by iron-catechol complexation. *Scientific Reports*, *10*, Article 8288. https://doi.org/10.1038/s41598-020-65171-1

Bittner, S. (2006). When quinones meet amino acids: Chemical, physical and biological consequences. *Amino Acids*, *30*(3), 205–224. https://doi.org/10.1007/s00726-005-0298-2

Blaut, M., Braune, A., Wunderlich, S., Sauer, P., Scheider, H., & Glatt, H. (2006). Mutagenicity of arbutin in mammalian cells after activation by human intestinal bacteria. *Food and Chemical Toxicology*, *44*, 1940–1947. https://doi.org/10.1016/j.fct.2006.06.015

Bock, A., Kieserling, H., Rohn, S., Steinhäuser, U., & Drusch, S. (2022). Impact of phenolic acid derivatives on β-lactoglobulin stabilized oil-water-interfaces. *Food Biophysics*, *17*(4), 508–522. https://doi.org/10.1007/s11483-022-09737-8

Bock, A., Rußbült, E., Barthel, L., Steinhäuser, U., Drusch, S., Rohn, S., & Kieserling, H. (2024). Co-adsorption and displacement behavior of phenolic acid derivatives at a β-lactoglobulin-stabilized oil–water interface and the interactions of both solutes. *Food Hydrocolloids*, *149*, Article 109578. https://doi.org/10.1016/j.foodhyd.2023.109578

Bohin, M. C., Vincken, J.-P., Van Der Hijden, H. T. W. M., & Gruppen, H. (2012). Efficacy of food proteins as carriers for flavonoids. *Journal of Agricultural and Food Chemistry*, *60*(16), 4136–4143. https://doi.org/10.1021/jf205292r

Bolton, J. L., Trush, M. A., Penning, T. M., Dryhurst, G., & Monks, T. J. (2000). Role of quinones in toxicology. *Chemical Research in Toxicology*, *13*(3), 135–215. https://doi.org/10.1021/tx9902082

Bongartz, V., Böttger, C., Wilhelmy, N., Schulze-Kaysers, N., Südekum, K. H., & Schieber, A. (2018). Protection of protein from ruminal degradation by alkali-induced oxidation of chlorogenic acid in sunflower meal. *Journal of Animal Physiology and Animal Nutrition*, *102*(1), 209–215. https://doi.org/10.1111/jpn.12729

Bongartz, V., Brandt, L., Gehrmann, M. L., Zimmermann, B. F., Schulze-Kaysers, N., & Schieber, A. (2016). Evidence for the formation of benzacridine derivatives in alkaline-treated sunflower meal and model solutions. *Molecules*, *21*(1), Article 91. https://doi.org/10.3390/molecules21010091

Bulaj, G., Kortemme, T., & Goldenberg, D. P. (1998). Ionization-reactivity relationships for cysteine thiols in polypeptides. *Biochemistry*, *37*(25), 8965–8972. https://doi.org/10.1021/bi973101r

Cao, W., Zhou, X., McCallum, N. C., Hu, Z., Ni, Q. Z., Kapoor, U., Heil, C. M., Cay, K. S., Zand, T., Mantanona, A. J., Jayaraman, A., Dhinojwala, A., Deheyn, D. D., Shawkey, M. D., Burkart, M. D., Rinehart, J. D., & Gianneschi, N. C. (2021). Unraveling the structure and function of melanin through synthesis. *Journal of the American Chemical Society*, *143*(7), 2622–2637. https://doi.org/10.1021/jacs.0c12322

Cao, Y., Xiong, Y. L., Cao, Y., & True, A. D. (2018). Interfacial properties of whey protein foams as influenced by preheating and phenolic binding at neutral pH. *Food Hydrocolloids*, *82*, 379–387. https://doi.org/10.1016/j.foodhyd.2018.04.020

Chen, N., Zhao, M., Chassenieux, C., & Nicolai, T. (2016). Structure of self-assembled native soy globulin in aqueous solution as a function of the concentration and the pH. *Food Hydrocolloids*, *56*, 417–424. https://doi.org/10.1016/j.foodhyd.2015.12.028

Chen, R., Wang, J. B., Zhang, X. Q., Ren, J., & Zeng, C. M. (2011). Green tea polyphenol epigallocatechin-3-gallate (EGCG) induced intermolecular cross-linking of membrane proteins. *Archives of Biochemistry and Biophysics*, *507*(2), 343–349. https://doi.org/10.1016/j.abb.2010.12.033

Chen, D., Zhu, X., Ilavsky, J., Whitmer, T., Hatzakis, E., Jones, O. G., & Campanella, O. H. (2021). Polyphenols weaken pea protein gel by formation of large aggregates with diminished noncovalent interactions. *Biomacromolecules*, *22*(2), 1001–1014. https://doi.org/10.1021/acs.biomac.0c01753

Cirkovic Velickovic, T. D., & Stanic-Vucinic, D. J. (2018). The role of dietary phenolic compounds in protein digestion and processing technologies to improve their antinutritive properties. *Comprehensive Reviews in Food Science and Food Safety*, *17*(1), 82–103. https://doi.org/10.1111/1541-4337.12320

Cladis, D. P., Weaver, C. M., & Ferruzzi, M. G. (2021). (Poly)phenol toxicity *in vivo* following oral administration: A targeted narrative review of (poly)phenols from green tea, grape and anthocyanine-rich extracts. *Phytotherapy Research*, *36*(1), 323–335. https://doi.org/10.1002/ptr.7323

Condict, L., Kaur, J., Hung, A., Ashton, J., & Kasapis, S. (2019). Combined spectroscopic, molecular docking and quantum mechanics study of β-casein and ferulic acid interactions following UHT-like treatment. *Food Hydrocolloids*, *89*, 351–359. https://doi.org/10.1016/j.foodhyd.2018.10.055

Corzo-Martínez, M., Soria, A. C., Belloque, J., Villamiel, M., & Moreno, F. J. (2010). Effect of glycation on the gastrointestinal digestibility and immunoreactivity of bovine beta-lactoglobulin. *International Dairy Journal*, *20*, 742–752. https://doi.org/10.1016/j.idairyj.2010.04.002

Croft, A. K., & Foley, M. K. (2008). Proline-rich proteins—Deriving a basis for residue-based selectivity in polyphenolic binding. *Organic and Biomolecular Chemistry*, *6*(9), 1594–1600. https://doi.org/10.1039/b800365c

Cusick, M. E., Klitgord, N., Vidal, M., & Hill, D. E. (2005). Interactome: Gateway into systems biology. *Human Molecular Genetics*, *14*(2), 171–181. https://doi.org/10.1093/hmg/ddi335

Dai, S., Lian, Z., Qi, W., Chen, Y., Tong, X., Tian, T., Lyu, B., Wang, M., Wang, H., & Jiang, L. (2022). Non-covalent interaction of soy protein isolate and catechin: Mechanism and effects on protein conformation. *Food Chemistry*, *384*, Article 132507. https://doi.org/10.1016/j.foodchem.2022.132507

Davis, P. J., & Williams, S. C. (1998). Protein modification by thermal processing. *Allergy*, *53*(s46), 102–105. https://doi.org/10.1111/j.1398-9995.1998.tb04975.x

de Oliveira, F. C., Coimbra, J. S. D. R., de Oliveira, E. B., Zuñiga, A. D. G., & Rojas, E. E. G. (2016). Food protein-polysaccharide conjugates obtained via the maillard reaction: A review. *Critical Reviews in Food Science and Nutrition*, *56*(7), 1108–1125. https://doi.org/10.1080/10408398.2012.755669

de Paula Rezende, J., Hudson, E. A., De Paula, H. M. C., Meinel, R. S., Da Silva, A. D., Da Silva, L. H. M., & dos Santos Pires, A. C. (2020). Human serum albumin-resveratrol complex formation: Effect of the phenolic chemical structure on the kinetic and thermodynamic parameters of the interactions. *Food Chemistry*,

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

307, Article 125514. https://doi.org/10.1016/j.foodchem.2019.125514

Drucker, C., Cicali, A. R., Roberts, A. M. P., Hughey, C. A., & Senger, L. W. (2023). Identification of alkaline-induced thiolyl-chlorogenic acid conjugates with cysteine and glutathione. *Food Chemistry*, *423*, Article 136267. https://doi.org/10.1016/j.foodchem.2023.136267

Ertop, M., & Bektaş, M. (2018). Enhancement of bioavailable micronutrients and reduction of antinutrients in foods with some processes. *Food and Health*, *4*(3), 159–165. https://doi.org/10.3153/fh18016

Ferrer-Gallego, R., Hernández-Hierro, J. M., Rivas-Gonzalo, J. C., & Escribano-Bailón, M. T. (2014). Sensory evaluation of bitterness and astringency sub-qualities of wine phenolic compounds: Synergistic effect and modulation by aromas. *Food Research International*, *62*, 1100–1107. https://doi.org/10.1016/j.foodres.2014.05.049

Foegeding, E. A., Luck, P. J., & Davis, J. P. (2006). Factors determining the physical properties of protein foams. *Food Hydrocolloids*, *20*(2-3), 284–292. https://doi.org/10.1016/j.foodhyd.2005.03.014

Fraga, C. G., Croft, K. D., Kennedy, D. O., & Tomás-Barberán, F. A. (2019). The effects of polyphenols and other bioactives on human health. *Food and Function*, *10*(2), 514–528. https://doi.org/10.1039/c8fo01997e

Friedman, M. (1996). Food browning and its prevention: An overview. *Journal of Agricultural and Food Chemistry*, *44*(3), 631–653. https://doi.org/10.1021/jf950394r

Galeb, H. A., Eichhorn, J., Harley, S., Robson, A. J., Martocq, L., Nicholson, S. J., Ashton, M. D., Abdelmohsen, H. A. M., Pelit, E., Baldock, S. J., Halcovitch, N. R., Robinson, B. J., Schacher, F. H., Chechik, V., Vercruysse, K., Taylor, A. M., & Hardy, J. G. (2023). Phenolic polymers as model melanins. *Macromolecular Chemistry and Physics*, *224*(14), Article 2300025. https://doi.org/10.1002/macp.202300025

Hasni, I., Bourassa, P., Hamdani, S., Samson, G., Carpentier, R., & Tajmir-Riahi, H. A. (2011). Interaction of milk α- And β-caseins with tea polyphenols. *Food Chemistry*, *126*(2), 630–639. https://doi.org/10.1016/j.foodchem.2010.11.087

He, Q., Lv, Y., & Yao, K. (2007). Effects of tea polyphenols on the activities of α-amylase, pepsin, trypsin and lipase. *Food Chemistry*, *101*(3), 1178–1182. https://doi.org/10.1016/j.foodchem.2006.03.020

Hellwig, M., & Henle, T. (2014). Baking, ageing, diabetes: A short history of the Maillard reaction. *Angewandte Chemie—International Edition*, *53*(39), 10316–10329. https://doi.org/10.1002/anie.201308808

Hofmann, T., Glabasnia, A., Schwarz, B., Wisman, K. N., Gangwer, K. A., & Hagerman, A. E. (2006). Protein binding and astringent taste of a polymeric procyanidin, 1,2,3,4,6-penta-O-galloyl-β-D-glucopyranose, castalagin, and grandinin. *Journal of Agricultural and Food Chemistry*, *54*(25), 9503–9509. https://doi.org/10.1021/jf062272c

Kumar, R., & Horigome, T. (1986). Fractionation, characterization, and protein-precipitating capacity of the condensed tannins from *Robinia pseudoacacia* L. leaves. *Journal of Agricultural and Food Chemistry*, *34*(3), 487–489. https://doi.org/10.1021/jf00069a029

Hu, B., Shen, Y., Adamcik, J., Fischer, P., Schneider, M., Loessner, M. J., & Mezzenga, R. (2018). Polyphenol-binding amyloid fibrils self-assemble into reversible hydrogels with antibacterial activity. *ACS Nano*, *12*(4), 3385–3396. https://doi.org/10.1021/acsnano.7b08969

Huang, H., Kwok, K. C., & Liang, H. (2004). Effects of tea polyphenols on the activities of soybean trypsin inhibitors and trypsin. *Journal of the Science of Food and Agriculture*, *84*(2), 121–126. https://doi.org/10.1002/jsfa.1610

Iacomino, M., Weber, F., Gleichenhagen, M., Pistorio, V., Panzella, L., Pizzo, E., Schieber, A., D'Ischia, M., & Napolitano, A. (2017). Stable benzacridine pigments by oxidative coupling of chlorogenic acid with amino acids and proteins: Toward natural product-based green food coloring. *Journal of Agricultural and Food Chemistry*, *65*(31), 6519–6528. https://doi.org/10.1021/acs.jafc.7b00999

Isaschar-Ovdat, S., & Fishman, A. (2018). Crosslinking of food proteins mediated by oxidative enzymes—A review. *Trends in Food Science and Technology*, *72*, 134–143. https://doi.org/10.1016/j.tifs.2017.12.011

Ishii, A. K., Pacioles, C. T., & Werem, L. (2021). Color and structural modifications of alkaline extracted sunflower protein concentrates and isolates using L-cysteine and glutathione. *Food Research International*, *147*, Article 110574. https://doi.org/10.1016/j.foodres.2021.110574

Jia, J., Gao, X., Hao, M., & Tang, L. (2017). Comparison of binding interaction between β-lactoglobulin and three common polyphenols using multi-spectroscopy and modeling methods. *Food Chemistry*, *228*, 143–151. https://doi.org/10.1016/j.foodchem.2017.01.131

Jia, W., Sethi, D. S., van der Goot, A. J., & Keppler, J. K. (2022). Covalent and non-covalent modification of sunflower protein with chlorogenic acid: Identifying the critical ratios that affect techno-functionality. *Food Hydrocolloids*, *131*, Article 107800. https://doi.org/10.1016/j.foodhyd.2022.107800

Jiang, J., Zhang, Z., Zhao, J., & Liu, Y. (2018). The effect of non-covalent interaction of chlorogenic acid with whey protein and casein on physicochemical and radical-scavenging activity of in vitro protein digests. *Food Chemistry*, *268*, 334–341. https://doi.org/10.1016/j.foodchem.2018.06.015

Jin, X. L., Wei, X., Qi, F. M., Yu, S. S., Zhou, B., & Bai, S. (2012). Characterization of hydroxycinnamic acid derivatives binding to bovine serum albumin. *Organic and Biomolecular Chemistry*, *10*(17), 3424–3431. https://doi.org/10.1039/c2ob25237f

Johnson, D. R., & Decker, E. A. (2015). The role of oxygen in lipid oxidation reactions: A review. *Annual Review of Food Science and Technology*, *6*, 171–190. https://doi.org/10.1146/annurev-food-022814-015532

Kallinich, C., Schefer, S., & Rohn, S. (2018). Analysis of protein-phenolic compound modifications using electrochemistry coupled to mass spectrometry. *Molecules*, *23*(2), 264–275. https://doi.org/10.3390/molecules23020264

Kanakis, C. D., Hasni, I., Bourassa, P., Tarantilis, P. A., Polissiou, M. G., & Tajmir-Riahi, H. A. (2011). Milk β-lactoglobulin complexes with tea polyphenols. *Food Chemistry*, *127*(3), 1046–1055. https://doi.org/10.1016/j.foodchem.2011.01.079

Karefyllakis, D., Altunkaya, S., Berton-Carabin, C. C., van der Goot, A. J., & Nikiforidis, C. V. (2017). Physical bonding between sunflower proteins and phenols: Impact on interfacial properties. *Food Hydrocolloids*, *73*, 326–334. https://doi.org/10.1016/j.foodhyd.2017.07.018

Keppler, J. K., van der Goot, A., & Reineccius, G. (2024). Hydrophobic/hydrophilic surface activity III: Interactions with other micro- and macromolecules. In J. P. D. Wanasundara, C. Schmitt, & B. P. Lamsal (Eds.), *Functionality of plant proteins: Properties, meth-*

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

ods of assessment, modifications and applications (pp. 197–221). Academic Press.

Keppler, J. K., Schwarz, K., & van der Goot, A. J. (2020). Covalent modification of food proteins by plant-based ingredients (polyphenols and organosulphur compounds): A commonplace reaction with novel utilization potential. *Trends in Food Science and Technology*, *101*, 38–49. https://doi.org/10.1016/j.tifs.2020.04.023

Keppler, J. K., Sönnichsen, F. D., Lorenzen, P. C., & Schwarz, K. (2014). Differences in heat stability and ligand binding among β-lactoglobulin genetic variants A, B and C using 1H NMR and fluorescence quenching. *Biochimica et Biophysica Acta—Proteins and Proteomics*, *1844*(6), 1083–1093. https://doi.org/10.1016/j.bbapap.2014.02.007

Kieserling, H., Pankow, A., Keppler, J. K., Wagemans, A. M., & Drusch, S. (2021). Conformational state and charge determine the interfacial film formation and film stability of β-lactoglobulin. *Food Hydrocolloids*, *114*, Article 106561. https://doi.org/10.1016/j.foodhyd.2020.106561

Kroll, J., Rawel, H. M., & Seidelmann, N. (2000). Physicochemical properties and susceptibility to proteolytic digestion of myoglobin-phenol derivatives. *Journal of Agricultural and Food Chemistry*, *48*(5), 1580–1587. https://doi.org/10.1021/jf991172m

Lambert, J. D., Sang, S., & Yang, C. S. (2007). Possible controversy over dietary polyphenols: Benefits vs risks. *Chemical Research in Toxicology*, *20*(4), 583–585. https://doi.org/10.1021/tx7000515

Le Bourvellec, C., & Renard, C.-M. (2012). Interactions between polyphenols and macromolecules: Quantification methods and mechanisms. *Critical Reviews in Food Science and Nutrition*, *52*, 213–248. https://doi.org/10.1080/10408398.2010.499808

Li, T., Li, X., Dai, T., Hu, P., Niu, X., Liu, C., & Chen, J. (2020). Binding mechanism and antioxidant capacity of selected phenolic acid—B-casein complexes. *Food Research International*, *129*, Article 108802. https://doi.org/10.1016/j.foodres.2019.108802

Li, T., Wang, L., Zhang, X., Yu, P., & Chen, Z. (2021). Complexation of rice glutelin fibrils with cyanidin-3-O-glucoside at acidic condition: Thermal stability, binding mechanism and structural characterization. *Food Chemistry*, *363*, Article 130367. https://doi.org/10.1016/j.foodchem.2021.130367

Li, X., Dai, T., Hu, P., Zhang, C., Chen, J., Liu, C., & Li, T. (2020). Characterization the non-covalent interactions between beta lactoglobulin and selected phenolic acids. *Food Hydrocolloids*, *105*, Article 105761. https://doi.org/10.1016/j.foodhyd.2020.105761

Liang, Y., & Were, L. (2020). Cysteine's effects on chlorogenic acid quinone induced greening and browning: Mechanism and effect on antioxidant reducing capacity. *Food Chemistry*, *309*, Article 125697. https://doi.org/10.1016/j.foodchem.2019.125697

Liu, J., Wang, Y., Tu, Z. C., Chen, W. M., & Yuan, T. (2021). Bovine β-lactoglobulin covalent modification by flavonoids: Effect on the allergenicity and human intestinal microbiota. *Journal of Agricultural and Food Chemistry*, *69*(24), 6820–6828. https://doi.org/10.1021/acs.jafc.1c02482

Lo Verde, C., Pacioles, C. T., Paterson, N., Chin, J., Owens, C. P., & Senger, L. W. (2023). Hydrolysis of chlorogenic acid in sunflower flour increases consumer acceptability of sunflower flour cookies by improving cookie color. *Journal of Food Science*, *88*(8), 3538–3550. https://doi.org/10.1111/1750-3841.16692

Lund, M. N. (2021). Reactions of plant polyphenols in foods: Impact of molecular structure. *Trends in Food Science and Technology*, *112*, 241–251. https://doi.org/10.1016/j.tifs.2021.03.056

Misnawi, J. S., Jamilah, B., & Nazamid, S. (2005). Changes in polyphenol ability to produce astringency during roasting of cocoa liquor. *Journal of the Science of Food and Agriculture*, *85*(6), 917–924. https://doi.org/10.1002/jsfa.1954

Moccia, F., Martín, M. Á., Ramos, S., Goya, L., Marzorati, S., DellaGreca, M., Panzella, L., & Napolitano, A. (2021). A new cyanine from oxidative coupling of chlorogenic acid with tryptophan: Assessment of the potential as red dye for food coloring. *Food Chemistry*, *348*, Article 129152. https://doi.org/10.1016/j.foodchem.2021.129152

Morzel, M., Canon, F., & Guyot, S. (2022). Interactions between salivary proteins and dietary polyphenols: Potential consequences on gastrointestinal digestive events. *Journal of Agricultural and Food Chemistry*, *70*(21), 6317–6327. https://doi.org/10.1021/acs.jafc.2c01183

Murata, M. (2022). Food chemistry and biochemistry of enzymatic browning. *Food Science and Technology Research*, *28*(1), 1–12. https://doi.org/10.3136/fstr.FSTR-D-21-00130

Narsimhan, G., & Xiang, N. (2018). Role of proteins on formation, drainage, and stability of liquid food foams. *Annual Review of Food Science and Technology*, *9*, 45–63. https://doi.org/10.1146/annurev-food-030216-030009

Nian, Y., Zhang, Y., Ruan, C., & Hu, B. (2022). Update of the interaction between polyphenols and amyloid fibrils. *Current Opinion in Food Science*, *43*, 99–106. https://doi.org/10.1016/j.cofs.2021.11.005

Nicolai, T., & Chassenieux, C. (2019). Heat-induced gelation of plant globulins. *Current Opinion in Food Science*, *27*, 18–22. https://doi.org/10.1016/j.cofs.2019.04.005

Noble, A. C. (2002). Astringency and Bitterness of Flavonoid Phenols. *ACS Symposium Series*, *825*, 192–201. https://doi.org/10.1021/bk-2002-0825.ch015

Ntone, E., Qu, Q., Gani, K. P., Meinders, M. B. J., Sagis, L. M. C., Bitter, J. H., & Nikiforidis, C. V. (2022). Sinapic acid impacts the emulsifying properties of rapeseed proteins at acidic pH. *Food Hydrocolloids*, *125*, Article 107423. https://doi.org/10.1016/j.foodhyd.2021.107423

Nucara, A., Maselli, P., Giliberti, V., & Carbonaro, M. (2013). Epicatechin-induced conformational changes in β-lactoglobulin B monitored by FT-IR spectroscopy. *SpringerPlus*, *2*(1), Article 661. https://doi.org/10.1186/2193-1801-2-661

Ozdal, T., Capanoglu, E., & Altay, F. (2013). A review on protein-phenolic interactions and associated changes. *Food Research International*, *51*(2), 954–970. https://doi.org/10.1016/j.foodres.2013.02.009

Pan, T., Wu, Y., He, S., Wu, Z., & Jin, R. (2022). Food allergenic protein conjugation with plant polyphenols for allergenicity reduction. *Current Opinion in Food Science*, *43*, 36–42. https://doi.org/10.1016/j.cofs.2021.10.002

Petzke, K. J., Schuppe, S., Rohn, S., Rawel, H. M., & Kroll, J. (2005). Chlorogenic acid moderately decreases the quality of whey proteins in rats. *Journal of Agricultural and Food Chemistry*, *53*(9), 3714–3720. https://doi.org/10.1021/jf048186z

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

Pessato, T. B., de Morais, F. P., de Carvalho, N. C., Figueira, A. C. M., Fernandes, L. G. R., Zollner, R. D. L., & Netto, F. M. (2018). Protein structure modification and allergenic properties of whey proteins upon interaction with tea and coffee phenolic compounds. *Journal of Functional Foods*, *51*, 121–129.

Pérot, M., Lupi, R., Guyot, S., Delayre-Orthez, C., Gadonna-Widehem, P., Thébaudin, J. Y., Bodinier, M., & Larré, C. (2017). Polyphenol interactions mitigate the immunogenicity and allergenicity of gliadins. *Journal of Agricultural and Food Chemistry*, *65*, 6442–6451.

Peyrot des Gachons, C., O'Keefe, A. J., Slade, L., & Beauchamp, G. K. (2021). Protein suppresses both bitterness and oleocanthal-elicited pungency of extra virgin olive oil. *Scientific Reports*, *11*(1), Article 11851. https://doi.org/10.1038/s41598-021-91046-0

Piber, M., & Koehler, P. (2005). Identification of dehydro-ferulic acid-tyrosine in rye and wheat: Evidence for a covalent cross-link between arabinoxylans and proteins. *Journal of Agricultural and Food Chemistry*, *53*(13), 5276–5284. https://doi.org/10.1021/jf050395b

Pires, M. A., Pastrana, L. M., Fucinõs, P., Abreu, C. S., & Oliveira, S. M. (2020). Sensorial perception of astringency: Oral mechanisms and current analysis methods. *Foods*, *9*(8), Article 1124. https://doi.org/10.3390/foods9081124

Poncet-Legrand, C., Edelmann, A., Putaux, J. L., Cartalade, D., Sarni-Manchado, P., & Vernhet, A. (2006). Poly(L-proline) interactions with flavan-3-ols units: Influence of the molecular structure and the polyphenol/protein ratio. *Food Hydrocolloids*, *20*(5), 687–697. https://doi.org/10.1016/j.foodhyd.2005.06.009

Prigent, S. V. E., Voragen, A. G. J., van Koningsveld, G. A., Baron, A., Renard, C. M. G. C., & Gruppen, H. (2009). Interactions between globular proteins and procyanidins of different degrees of polymerization. *Journal of Dairy Science*, *92*(12), 5843–5853. https://doi.org/10.3168/jds.2009-2261

Prigent, S. V. E., Gruppen, H., Visser, A. J. W. G., Van Koningsveld, G. A., De Jong, G. A. H., & Voragen, A. G. J. (2003). Effects of non-covalent interactions with 5-O-caffeoylquinic acid (chlorogenic acid) on the heat denaturation and solubility of globular proteins. *Journal of Agricultural and Food Chemistry*, *51*(17), 5088–5095. https://doi.org/10.1021/jf021229w

Pu, P., Zheng, X., Jiao, L., Chen, L., Yang, H., Zhang, Y., & Liang, G. (2021). Six flavonoids inhibit the antigenicity of β-lactoglobulin by noncovalent interactions: A spectroscopic and molecular docking study. *Food Chemistry*, *339*, Article 128106. https://doi.org/10.1016/j.foodchem.2020.128106

Queiroz, C., Mendes Lopes, M. L., Fialho, E., & Valente-Mesquita, V. L. (2008). Polyphenol oxidase: Characteristics and mechanisms of browning control. *Food Reviews International*, *24*(4), 361–375. https://doi.org/10.1080/87559120802089332

Rawel, H. M., Kroll, J., & Riese, B. (2000). Reactions of chlorogenic acid with lysozyme: Physicochemical characterization and proteolytic digestion of the derivatives. *Journal of Food Science*, *65*(6), 1091–1098. https://doi.org/10.1111/j.1365-2621.2000.tb09424.x

Rawel, H. M., Kroll, J., & Hohl, U. C. (2001). Model studies on reactions of plant phenols with whey proteins. *Food/Nahrung*, *45*(2), 72–81.

Rawel, H. M., Czajka, D., Rohn, S., & Kroll, J. (2002). Interactions of different phenolic acids and flavonoids with soy proteins. *International Journal of Biological Macromolecules*, *30*(3–4), 137–150. https://doi.org/10.1016/S0141-8130(02)00016-8

Rawel, H. M., Kroll, J., & Rohn, S. (2001). Reactions of phenolic substances with lysozyme—Physicochemical characterisation and proteolytic digestion of the derivatives. *Food Chemistry*, *72*(1), 59–71. https://doi.org/10.1016/S0308-8146(00)00206-5

Rawel, H. M., Meidtner, K., & Kroll, J. (2005). Binding of selected phenolic compounds to proteins. *Journal of Agricultural and Food Chemistry*, *53*(10), 4228–4235. https://doi.org/10.1021/jf0480290

Rawel, H. M., & Rohn, S. (2010). Nature of hydroxycinnamate-protein interactions. *Phytochemistry Reviews*, *9*(1), 93–109. https://doi.org/10.1007/s11101-009-9154-4

Richard, T., Vitrac, X., Merillon, J. M., & Monti, J. P. (2005). Role of peptide primary sequence in polyphenol-protein recognition: An example with neurotensin. *Biochimica et Biophysica Acta—General Subjects*, *1726*(3), 238–243. https://doi.org/10.1016/j.bbagen.2005.07.017

Riihimäki, L. H., Vainio, M. J., Heikura, J. M. S., Valkonen, K. H., Virtanen, V. T., & Vuorela, P. M. (2008). Binding of phenolic compounds and their derivatives to bovine and reindeer β-lactoglobulin. *Journal of Agricultural and Food Chemistry*, *56*(17), 7721–7729. https://doi.org/10.1021/jf801120a

Rohn, S. (2014). Possibilities and limitations in the analysis of covalent interactions between phenolic compounds and proteins. *Food Research International*, *65*, 13–19. https://doi.org/10.1016/j.foodres.2014.05.042

Rohn, S., Petzke, K. J., Rawel, H. M., & Kroll, J. (2006). Reactions of chlorogenic acid and quercetin with a soy protein isolate—Influence on the in vivo food protein quality in rats. *Molecular Nutrition and Food Research*, *50*(8), 696–704. https://doi.org/10.1002/mnfr.200600043

Rossetti, D., Yakubov, G. E., Stokes, J. R., Williamson, A. M., & Fuller, G. G. (2008). Interaction of human whole saliva and astringent dietary compounds investigated by interfacial shear rheology. *Food Hydrocolloids*, *22*(6), 1068–1078. https://doi.org/10.1016/j.foodhyd.2007.05.014

Rubino, M. I., Arntfield, S. D., Nadon, C. A., & Bernatsky, A. (1996). Phenolic protein interactions in relation to the gelation properties of canola protein. *Food Research International*, *29*(7), 653–659. https://doi.org/10.1016/S0963-9969(97)89643-3

Sabir, M. A., Sosulski, F. W., & Finlayson, A. J. (1974). Chlorogenic acid-protein interactions in sunflower. *Journal of Agricultural and Food Chemistry*, *22*(4), 575–578. https://doi.org/10.1021/jf60194a052

Sagis, L. M., & Yang, J. (2022). Protein-stabilized interfaces in multiphase food: Comparing structure-function relations of plant-based and animal-based proteins. *Current Opinion in Food Science*, *43*, 53–60. https://doi.org/10.1016/j.cofs.2021.11.003

Santarcangelo, A., Schulze-Kaysers, N., & Schieber, A. (2024). Improved generation, physicochemical characteristics, and food application studies of a red colorant obtained from oxidative coupling of chlorogenic acid and tryptophan. *Foods*, *13*(5), Article 686. https://doi.org/10.3390/foods13050686

Santarcangelo, A., Weber, F., Kehraus, S., Dickschat, J. S., & Schieber, A. (2023). Generation and structure elucidation of a red colorant formed by oxidative coupling of chlorogenic acid and tryptophan. *Food Chemistry*, *425*, Article 136473. https://doi.org/10.1016/j.foodchem.2023.136473

Scalbert, A., Manach, C., Morand, C., Rémésy, C., & Jiménez, L. (2007). Dietary polyphenols and the prevention of diseases. *Crit-*

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

ical Reviews in Food Science and Nutrition, *45*(4), 287–306. https://doi.org/10.1080/1040869059096

Schieber, A. (2018). Reactions of quinones—Mechanisms, structures, and prospects for food research. *Journal of Agricultural and Food Chemistry*, *66*(50), 13051–13055. https://doi.org/10.1021/acs.jafc.8b05215

Schild, K., Sönnichsen, F. D., Martin, D., Garamus, V. M., Van der Goot, A. J., Schwarz, K., & Keppler, J. K. (2023). Unraveling the effects of low protein-phenol binding affinity on the structural properties of beta-lactoglobulin. *Food Chemistry*, *426*, Article 136496. https://doi.org/10.1016/j.foodchem.2023.136496

Selinheimo, E., Lampila, P., Mattinen, M., & Buchert, J. (2008). Formation of protein—Oligosaccharide conjugates by laccase and tyrosinase. *Journal of Agricultural and Food Chemistry*, *56*, 3118–3128. https://doi.org/10.1021/jf0730791

Shahidi, F., & Dissanayaka, C. S. (2023). Phenolic-protein interactions: Insight from in-silico analyses—A review. *Food Production, Processing and Nutrition*, *5*, Article 2. https://doi.org/10.1186/s43014-022-00121-0

Singh, H. (2011). Aspects of milk-protein-stabilised emulsions. *Food Hydrocolloids*, *25*(8), 1938–1944. https://doi.org/10.1016/j.foodhyd.2011.02.022

Sinha, M., Sachan, D. K., Bhattacharya, R., Singh, P., & Parthasarathi, R. (2022). ToxDP2 Database: Toxicity prediction of dietary polyphenols. *Food Chemistry*, *370*, Article 131350. https://doi.org/10.1016/j.foodchem.2021.131350

Skrt, M., Benedik, E., Podlipnik, Č., & Ulrih, N. P. (2012). Interactions of different polyphenols with bovine serum albumin using fluorescence quenching and molecular docking. *Food Chemistry*, *135*(4), 2418–2424. https://doi.org/10.1016/j.foodchem.2012.06.114

Soares, S., Silva, M. S., Garcia-Estevez, I., Großmann, P., Bras, N., Brandao, E., Mateus, N., Freitas, V., Behrens, M., & Meyerhof, W. (2018). Human bitter taste receptors are activated by different classes of polyphenols. *Journal of Agricultural and Food Chemistry*, *66*, 8814–8823. https://doi.org/10.1021/acs.jafc.8b03569

Sosulski, F. (1979). Organoleptic and nutritional effects of phenolic compounds on oilseed protein products: A review. *Journal of the American Oil Chemists' Society*, *56*(8), 711–715. https://doi.org/10.1007/BF02663047

Stojadinovic, M., Radosavljevic, J., Ognjenovic, J., Vesic, J., Prodic, I., Stanic-Vucinic, D., & Cirkovic Velickovic, T. (2013). Binding affinity between dietary polyphenols and β-lactoglobulin negatively correlates with the protein susceptibility to digestion and total antioxidant activity of complexes formed. *Food Chemistry*, *136*(3–4), 1263–1271. https://doi.org/10.1016/j.foodchem.2012.09.040

Sui, X., Sun, H., Qi, B., Zhang, M., Li, Y., & Jiang, L. (2018). Functional and conformational changes to soy proteins accompanying anthocyanins: Focus on covalent and non-covalent interactions. *Food Chemistry*, *245*, 871–878. https://doi.org/10.1016/j.foodchem.2017.11.090

Tagliazucchi, D., Verzelloni, E., & Conte, A. (2005). Effect of some phenolic compounds and beverages on pepsin activity during simulated gastric digestion. *Journal of Agricultural and Food Chemistry*, *53*(22), 8706–8713. https://doi.org/10.1021/jf058074n

Tan, C., Xu, Q. Da, Chen, N., He, Q., Sun, Q., & Zeng, W. C. (2022). Cross-linking effects of EGCG on myofibrillar protein from common carp (Cyprinus carpio) and the action mechanism. *Journal of Food Biochemistry*, *46*(12), Article e14416. https://doi.org/10.1111/jfbc.14416

Tantoush, Z., Apostolovic, D., Kravic, B., Prodic, I., Mihajlovic, L., Stanic-Vucinic, D., & Cirkovic Velickovic, T. (2012). Green tea catechins of food supplements facilitate pepsin digestion of major food allergens, but hampers their digestion if oxidized by phenol oxidase. *Journal of Functional Foods*, *4*(3), 650–660. https://doi.org/10.1016/j.jff.2012.04.006

Tarahi, M., Gharagozlou, M., Niakousari, M., & Hedayati, S. (2024). Protein-chlorogenic acid interactions: Mechanisms, characteristics, and potential food applications. *Antioxidants*, *13*, Article 777. https://doi.org/10.3390/antiox13070777

Tsai, P. J., & She, C. H. (2006). Significance of phenol-protein interactions in modifying the antioxidant capacity of peas. *Journal of Agricultural and Food Chemistry*, *54*(22), 8491–8494. https://doi.org/10.1021/jf061475y

von Staszewski, M., Pizones Ruiz-Henestrosa, V. M., & Pilosof, A. M. R. (2014). Green tea polyphenols-β-lactoglobulin nanocomplexes: Interfacial behavior, emulsification and oxidation stability of fish oil. *Food Hydrocolloids*, *35*, 505–511. https://doi.org/10.1016/j.foodhyd.2013.07.008

Vreeke, G. J. C., Lubbers, W., Vincken, J.-P., & Wierenga, P. A. (2022). A method to identify and quantify the complete peptide composition in protein hydrolysates. *Analytica Chimica Acta*, *1201*, Article 339616. https://doi.org/10.1016/j.aca.2022.339616

Wildermuth, S. R., Young, E. E., & Were, L. M. (2016). Chlorogenic acid oxidation and its reaction with sunflower proteins to form green-colored complexes. *Comprehensive Reviews in Food Science and Food Safety*, *15*(5), 829–843. https://doi.org/10.1111/1541-4337.12213

Wodak, S. J., Vlasblom, J., Turinsky, A. L., & Pu, S. (2013). Protein-protein interaction networks: The puzzling riches. *Current Opinion in Structural Biology*, *23*(6), 941–953. https://doi.org/10.1016/j.sbi.2013.08.002

Xu, Z., Shan, G., Hao, N., Li, L., Lan, T., Dong, Y., Wen, J., Tian, R., Zhang, Y., Jiang, L., & Sui, X. (2022). Structure remodeling of soy protein-derived amyloid fibrils mediated by epigallocatechin-3-gallate. *Biomaterials*, *283*, Article 121455. https://doi.org/10.1016/j.biomaterials.2022.121455

Yabuta, G., Koizumi, Y., Namiki, K., Hida, M., & Namiki, M. (2001). Structure of green pigment formed by the reaction of caffeic acid esters (or chlorogenic acid) with a primary amino compound. *Bioscience, Biotechnology and Biochemistry*, *65*(10), 2121–2130. https://doi.org/10.1271/bbb.65.2121

Yan, X., Chu, Y., Hang, J., Zhang, S., Yu, P., Gong, X., Zeng, Z., & Chen, L. (2024). Enhancing emulsifying properties of lentil protein fibrils through EGCG mediation and the mechanism study. *Food Hydrocolloids*, *153*, Article 109972. https://doi.org/10.1016/j.foodhyd.2024.109972

Yang, J., Lamochi Roozalipour, S. P., Berton-Carabin, C. C., Nikiforidis, C. V., van der Linden, E., & Sagis, L. M. C. (2021). Air-water interfacial and foaming properties of whey protein—Sinapic acid mixtures. *Food Hydrocolloids*, *112*, Article 106467. https://doi.org/10.1016/j.foodhyd.2020.106467

Yu, J., Mi, Y., & Ji, S. (2016). In vitro evaluating the influence of grape seed polyphenol extract on the digestibility of macronutrients. *Journal of Health Science*, *4*(4), 167–176. https://doi.org/10.17265/2328-7136/2016.04.001

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License

Zee, B. M., & Garcia, B. A. (2012). Discovery of lysine post-translational modifications through mass spectrometric detection. *Bone*, *52*, 147–163. https://doi.org/10.1042/bse0520147

Zhang, Q., Cheng, Z., Wang, Y., & Fu, L. (2021). Dietary protein-phenolic interactions: Characterization, biochemical-physiological consequences, and potential food applications. *Critical Reviews in Food Science and Nutrition*, *61*(21), 3589–3615. https://doi.org/10.1080/10408398.2020.1803199

Zondlo, N. J. (2012). Tunable CH /π interactions. *Accounts of Chemical Research*, *46*(4), 1039–1049.

How to cite this article: Kieserling, H., Bruijn, W. J. C., Keppler, J., Yang, J., Sagu, S. T., Güterbock, D., Rawel, H., Schwarz, K., Vincken, J.-P., Schieber, A., & Rohn, S. (2024). Protein–phenolic interactions and reactions: Discrepancies, challenges, and opportunities. *Comprehensive Reviews in Food Science and Food Safety*, *23*, e70015. https://doi.org/10.1111/1541-4337.70015

15414337, 2024, 5, Downloaded from https://ift.onlinelibrary.wiley.com/doi/10.1111/1541-4337.70015 by Wageningen University And Research Facilitair Bedrijf, Wiley Online Library on [19/09/2024]. See the Terms and Conditions (https://onlinelibrary.wiley.com/terms-and-conditions) on Wiley Online Library for rules of use; OA articles are governed by the applicable Creative Commons License