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# Lipid oxidation in food emulsions: a review dedicated to the role of the interfacial area

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In this review, we focus on the role of the interface in lipid oxidation in food emulsions. Mostly, results from this field are a reflection of the effects caused by reaction kinetics and mass transfer, which complicates interpretation. In general, the oil–water interface is the location of initiation of oxidation reactions, while components present there, and in the continuous phase, directly or indirectly affect the reaction. Smaller droplets are expected to oxidize faster, but this can be counteracted by components purposely positioned at the interface or added to the bulk phase. Recent simulation progress is expected to be instrumental in distinguishing these effects, and guides stable emulsion design.

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## Introduction

### Lipid oxidation

Lipid oxidation in foods is a major problem, as it causes degradation of product quality that alters the textural properties, and also affects the color and nutritional value of food product. In foods, unsaturated fatty acids that are part of mostly triacylglycerols [1] and phospholipids ultimately decompose into volatile compounds that produce rancidity with off-odors. The susceptibility of fatty acids to lipid oxidation increases with the degree of unsaturation due to progressively lower bond dissociation energies of methylene-interrupted carbons [2]. This makes following the latest Dietary Guidelines [3,4]

that promote the use of polyunsaturated fatty acids as a healthier source of fats for consumers [5] progressively difficult. It is notable that oxidation reactions are strongly affected by temperature, and that effects found at high temperature are not necessarily relevant for shelf life at room temperature (different oxygen solubility, partial pressure, and/or side product formation, for example, through caramelization and Maillard reactions), although it is understandable that for time considerations, accelerated shelf-life tests are performed [6].

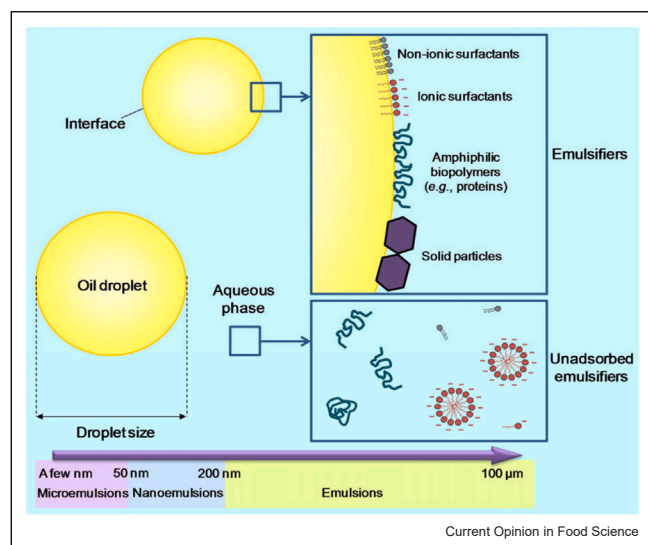
Lipid and also protein oxidation that are expected to be intertwined in protein-stabilized emulsions, are both radical reactions that include reactive oxygen species [7]. These reactions can be catalyzed by, for example, metal ions that cause the development of a broad range of oxidation products, such as hydroperoxides (primary oxidation product) and secondary oxidation products such as aldehydes, ketones, and alcohols. Formation of peptides, aggregation, and modification of side chains can occur as a result of protein oxidation [8,9], which in turn may cause changes in protein surface hydrophobicity [10,11].

### Food emulsions

During emulsion formation, the surface area created can be as high as thousands of m<sup>2</sup> per gram of dispersed oil [12]. It has been suggested, and to some extent proven, that oxidation reactions are initiated at the interface, which makes emulsion highly sensitive systems, especially when highly unsaturated fats are used [13]. For mayonnaise, for example, the appearance of aldehydes is considered a measure of oxidative stability [14]. To combat this, antioxidants can be used, and depending on the oil matrix, the effects can be very different, and this would also hold for the emulsification method used that will influence among others the interfacial composition [15,16]. In the work done in the group of Pierre Ville-neuve, the cutoff effect [17] was reported, with the chain length of antioxidant determining its location and therewith its effect on oxidation. It is good to point out that if antioxidants are very hydrophobic, they would partition into the oil droplet, and away from the interface, thus making them rather ineffective in preventing lipid oxidation, as was recently confirmed for dried emulsions [18].

The emulsification method used [19,20], as well as the ingredients, influences droplet interface stabilization and

Figure 1



Schematic representation of the main physical properties, compartments, and possible components of oil-in-water emulsions. (Picture is taken from presentation and used with permission of the presenter).

droplet coalescence through dynamic effects [21] that can occur in the submillisecond time range [22–24]. When cavitation occurs, for example, during high-pressure homogenization, this can induce radical formation, thus initiating lipid oxidation. Physical emulsion destabilization (flocculation, coalescence) [25] may be amplified when oxidation reactions influence the composition of the interface.

## Lipid oxidation at various levels

### Emulsion composition

Figure 1 shows the various size scales of an emulsion, going from droplets to the interfacial region, and components in the continuous phase, that may be present as micelles. The interfacial layer may be thin (a few nm), but for relatively small droplets (size  $\sim 0.1 \mu\text{m}$ ), it holds considerable volume [26]. In emulsions, molecules partition between the three regions based on their polarity. Nonpolar molecules are mostly present in the oil, while polar molecules primarily locate in the aqueous region; molecules of intermediate polarity would be more prominently present in the interfacial region [27]. In the literature, this has been linked to the effectiveness of antioxidants, that when positioned in the interface, are much more effective [28], and for which the cutoff concept was proposed [29,30].

A logP value (the partitioning coefficient in an octane/water two-phase system) can be instrumental in determining where a component would reside preferentially, although it is good to keep in mind that this

is an indication for what would happen in an emulsion and not a prediction since, for example, vegetable oil and water would have more extreme partitioning due to the higher hydrophobicity of the oil compared with octane, and the effects would depend on the hydrophobicity of the component considered. Because of equilibrium considerations, (very) small amounts would be present in all phases, and when highly reactive, they may still be influential. Furthermore, the partial pressure is relevant for volatile components, which may make them partition to the gas phase (head space), and thus no longer participate in the oxidation reaction cascade. Both the partial pressure and logP values allow comparison of component behavior in an unbiased way, although given the complexity of emulsions, this may not cover all relevant effects, as is described in the next section for the interfacial layer and the bulk phase.

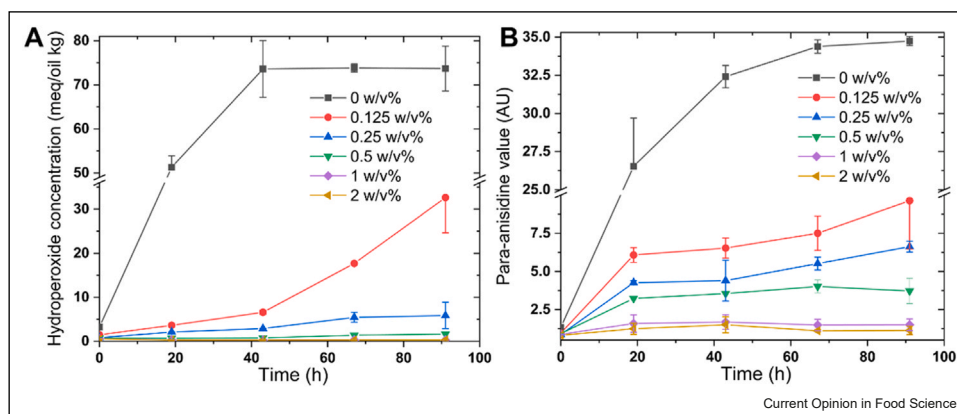
### Interface effects

Various routes have been described in the literature for initiation of lipid oxidation [31], such as spontaneous abstraction of a hydrogen from an unsaturated fatty acid, or reinitiation through decomposition of hydroperoxides by, for example, trace metal ions [32], these reactions are both expected to be a function of the amount of interface available as well as its composition. It is good to mention that for the latter effect, this implies that the amount of hydroperoxides that are initially present will influence the course of reaction as illustrated in the recent modeling study by Schroën and Berton-Carabin [33]. For both types of initiation, there is a central role for metal ions that catalyze the reaction, and since these ions partition between interface and bulk water phase, it is clear that affinity for these components by other components present in the bulk phase would influence the amount of a catalyst present at the interface, and thus oxidation [28], as described in greater detail in the next section.

### Effects occurring in the bulk water phase

In various studies on oxidation in emulsions, the reaction is carried out in the presence of a combination of a metal catalyst, and chelating components such as Ethylenediaminetetraacetic acid (EDTA). Metal chelation inhibits decomposition of hydroperoxides by preventing cationic pro-oxidants [34], such as metals, to reach the hydroperoxides near the droplet surface and thus reduce oxidation. Besides, it is known that proteins may have metal-binding capacity above their isoelectric point [9,35], and for  $\beta$ -casein and its hydrolysates, this was related to the presence of phosphoryl groups that remain negatively charged even at low pH. It has been reported that casein that remains in the bulk phase can protect the lipid phase against oxidation in this way [34], as was reported for other proteins. Furthermore, proteins can scavenge free radicals originating from lipid oxidation [35–37], thus slowing oxidation [38] in casein-,

Figure 2



Hydroperoxide concentration (a) and para-anisidine values (b) in WPI-stabilized emulsions supplemented with 0–2 w/v% high-molecular-weight coffee melanoidin fraction incubated at 40°C for four days (submitted work).

NaCas- [39,40], and potato protein-stabilized emulsions [41,42].

In earlier work, we found that soy protein isolates (SPI) at neutral pH [43] had better antioxidant power than casein, which in turn was more effective than whey protein isolate (WPI). The antioxidant ability of SPI and WPI was assigned to the sulfhydryl group of the proteins [35], and more in general, the tendency of amino acid residues to scavenge free radicals depending on the tertiary structure of the proteins; when insufficiently exposed, they will not be able to participate in free radical scavenging [44]. Effects both in the oil phase and in the water phase can be expected to play a role, also given the interconnectedness of protein and lipid oxidation [45]. Tween 20 has been reported to change protein conformation and increase accessibility of certain amino acid residues for radical scavenging [43,46].

It is important to point out that components used in emulsion formulations may undergo reactions, for example, Maillard reactions when given a heat treatment. This will affect the capacity of these proteins to influence lipid oxidations in different ways. Very recently, we showed [47] that glycosylation of SPI can lead to a direct effect on oxidation at the interface, but also indirectly when added to the bulk phase of a whey protein-stabilized emulsion of which the oxidative stability was remarkably improved [48]. In submitted work, (see Figure 2), we have used coffee melanoidin fractions, and found remarkably improved oxidative stability in emulsions when added post emulsification to the bulk phase.

Lately, papers and reviews have been published that are dedicated to the effect that micellar structures may have on lipid oxidation, for example, [32,49]. It is suggested that (enhanced) transfer of the oxidation products takes

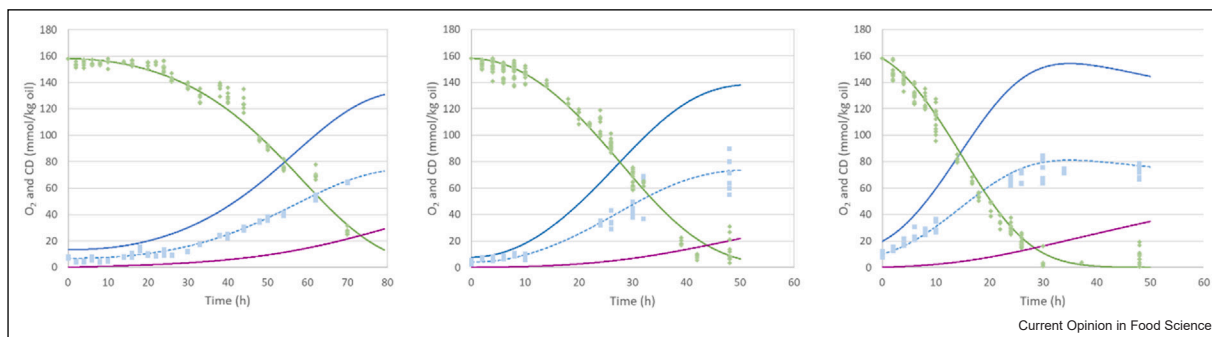
place through micelles present in the bulk phase, which would allow oxidized droplets to ‘contaminate’ clean ones. Mostly, these considerations are still theoretical in nature, but interesting. Transfer between droplets has been visualized for mayonnaise using Confocal Laser Scanning Microscopy (CLSM) [50], although the direct contact between the droplet may have led to direct transfer, without the need of any micellar transfer. In a submitted paper, we investigated transfer of primary and secondary oxidation products, and found that no noticeable transfer of primary products takes place within the timespan of the experiments (two weeks) when connected to the triglyceride, and only limitedly for short-chain secondary oxidation products.

#### Modeling oxidation in food emulsions

To capture the complexity of the oxidation reaction, for example, an exponential approach after a lag phase [51] was used, that can be quantified by using a Gompertz model [14], and a pseudo-phase kinetic model was proposed to describe the effect of antioxidants [37,52]. Very recently, we have published a first-order reaction approach, and have obtained good agreement both for protein- and surfactant-stabilized emulsions; [33] see Figure 3. In that way, we were able to distinguish initiation of oxidation between two types of emulsions: for surfactant-stabilized emulsions that are most probably through reinitiation by decomposition of hydroperoxides, while for protein-stabilized emulsions, this is expected to be mostly through spontaneous radical formation, that is the result of the interplay between protein and lipid oxidation.

When using the model to predict different scenarios, quite interestingly, oxidation in emulsions of different size stabilized by Tween 20 and Tween 80 could be unified by taking the interfacial area into account in the

Figure 3



Experimentally determined amounts of consumed oxygen (green diamonds), and produced conjugated dienes (blue squares), and the predicted values for oxygen concentration (green), conjugated dienes (blue dashed line), total hydroperoxides, and total secondary products (purple) in Tween 80- (left), bovin serum albumin- (middle), and  $\beta$ -lactoglobulin-stabilized emulsions (right) [33].

radical initiation reaction rate constant when starting from hydroperoxides. This is an indication that in these very well-controlled and mixed emulsions, the surface area may determine the oxidation rate, but this is not confirmed yet, and part of ongoing work in various labs focusing on the effect of mass transfer, subphase composition, effects of bulk components, and so on, on lipid oxidation. Furthermore, the effect of the initial amount of hydroperoxides could be calculated using the model, and that showed that this greatly affected the oxidation rate in surfactant-stabilized emulsions (see Graphical Abstract). Hydroperoxide formation is of course related to radical formation, and thus propagation of the reaction, which explains the model result, but it is quite remarkable that initial hydroperoxide concentration is not standardly taken into account when explaining differences in oxidation in emulsions. Since oxidation is an intertwined effect with protein oxidation, also this should be considered as an explanation for the differences found, but in practice, this is not done.

In general, oxidation is strongly affected by temperature, and in principle, these effects can be covered by the previously mentioned model [33] when making the reaction rate constants temperature-dependent. As mentioned previously, the effects found at high temperatures are not necessarily relevant for shelf life at room temperature, and here, we give as an additional argument that the temperature dependency of the reactions is simply different, leading to a shift in the ratio primary versus secondary oxidation products, which makes comparison between temperatures inherently more difficult.

### Innovative approaches for food emulsion design

Since lipid oxidation is initiated at the oil/water interface, it is relevant to design the emulsion and position components in such a way that they can optimally

contribute to product stability [26]. In the previous section, this was already illustrated by discussing the action of proteins and Maillard reaction products at the interface and in the bulk of the emulsions.

An interesting approach is to put antioxidants in the interface by the use of so-called Pickering particles [53]. Although there is a lot of discussion about what is a Pickering emulsion when claimed in the food field (in our opinion, mostly mixed interfaces are created), the use of particles is relevant since they can nest at the interface practically irreversibly and thus promote the physical stability of emulsions for a long period [54–58] even up to several years [59]. The amount of particles needed follows from equations as presented, for example, [55] for the ideal case that all particles are in the interface.

$$D = \frac{4C\rho_p d_p V_d}{m_p}$$

Where  $D$  is the droplet size,  $C$  is the degree of coverage,  $\rho_p$  is the particle density ( $\text{kg/m}^3$ ),  $d_p$  is the particle diameter (m),  $V_d$  is the volume of dispersed phase ( $\text{m}^3$ ), and  $m_p$  is the mass of particles (kg). It has been shown that when antioxidant-containing solid-fat particles are used to stabilize an emulsion, this leads to both enhanced physical and oxidative stability. It is expected that capture of the antioxidant needs to be improved compared with Schroder et. al., [60], since the antioxidant slowly leaches from the solid lipid particles to the oil phase [61]. Still, it is an interesting concept that may also be realized through the use of natural particles containing antioxidants [62,63].

### Conclusions

Various factors related to the interface, and to the bulk phase, contribute to oxidative stability of emulsions relevant for food and many other fields. Quantification of

these effects is far from trivial, and it is expected that some new developments, especially those that allow evaluation of the cascade of oxidation reactions through modeling, will be instrumental in pinpointing the relevance of the constituent reactions, and the role of the droplet size. In itself, it is logical that with more interface present, the reaction would proceed much faster, but this would also greatly depend, for example, on the presence of components that may influence the reaction by, for example, radical scavenging or metal chelation (by proteins or other agents). A relatively new development is the use of glycosylated proteins in the bulk phase, and also that of particles with antioxidant capacity at the interface. Both options lead to emulsions with considerably higher oxidative stability, and are interesting leads for making emulsions inherently more stable, and thus contribute to reducing food waste.

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### CRedit authorship contribution statement

**Asif Aslam:** Writing – original draft, Conceptualization;  
**Karin Schroën:** Writing – review & editing, Conceptualization.

### Conflict of interest statement

None declared.

### Data availability

No data were used for the research described in the article.

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