Dynamic Release of Diacetyl from Liquid Gelatin in the Headspace

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The effect of gelatin concentration and stirring rate on the dynamic release of diacetyl was investigated, using headspace analysis for the duration of the experiment. Both liquid and headspace were stirred in a sealed system. An increase in gelatin concentration was shown to affect the initial release rate, as defined by the amount of diacetyl released over the first few minutes of the experiment. It also affected the equilibrium partitioning of the diacetyl between the aqueous concentration of the gelatin solution and the headspace, which indicates binding between diacetyl and gelatin. An increase in stirring rate of the solutions of the same composition showed an increase in the release rate of diacetyl, while an increase in viscosity showed a decrease in the release rate. Mathematical models previously developed to describe flavor release from aqueous solutions containing flavor-binding polymers (Harrison, M.; Hills, B. P. *J. Agric. Food Chem*. **1997**, *45*, 1883−1890) were used to interpret the data. Analysis of the time-dependent release data confirmed that transfer of diacetyl flavor molecules across the liquid−gas interface is rate limiting and is adequately described by the penetration theory of mass transfer.

Keywords: *Flavor release; gelatin; diacetyl; modeling*

INTRODUCTION

The perception of a flavor is an enjoyable aspect of eating and therefore an important parameter of consumer acceptability of a food. Hence, the study of the interactions of flavor with a food matrix and their effects on the release and perception of flavor is an important area of study. Many sensory studies have been conducted, but comparison with instrumental data of quantitative and qualitative flavor composition remains difficult to interpret, and predictions regarding the sensory properties of a food from the compositional data cannot easily be made. One important difference between a sensory assessment and an instrumental measurement is that during eating the flavor is released and perceived during a short time span, while the food is generally broken down by chewing, mixed with saliva, and in certain cases in part melted. The flavor travels via an airflow to the olfactory epithelium, where receptor proteins interact with the flavor compounds and a chain of events is started leading to our perception of a flavor. The concentration of the flavor stimulus from a complex food is difficult to determine, due to its dependence on the food matrix. Flavor perception depends on the composition of the food matrix and the breakdown processes during eating; these processes will have to occur in a short time, typically <2 min. The factors influencing the release and perception of flavors are discussed in a recent review (Bakker, 1995). It is likely that the equilibrium condition as defined by the partition coefficient, which has been the subject of many investigations [see review by Bakker (1995)] is not attained during eating but that the rate of release of flavor is especially important for interpreting the perception of flavor.

Hence, instrumental measurements to determine the rates of release may well be invaluable to determine the amount of flavor available for sensory perception. Darling et al. (1986) suggested that when the applied stimulus for sensory evaluation is a food, it is important to know the interaction between the flavor and the structure/composition of a food to define the applied stimulus. One of the first studies monitoring the headspace concentration continuously by mass spectrometer was done by Lee (1986), showing an effect of fat saturation on the release of diacetyl. Salvador et al. (1994) were among the first authors to report release rates of flavors from emulsions, defined as the amount of flavor released as a function of time. In a rather simple setup they demonstrated an effect of the structure of the emulsion on the release rate of flavor as well as effects of stirring rates and air renewal. A number of approaches have been published since to break down the food to determine the release of flavor under conditions mimicking the types of breakdown and mixing occurring during eating (Bakker, 1995). For example, Roberts and Acree (1995) used a food blender incorporating purge and trap to simulate the release of aroma in the mouth.

A previous study (Roberts et al., 1996) using a mass spectrometer to monitor continuous release curves showed an effect of thickener on the release of selected volatiles. These authors reported that an increase in carboxymethylcellulose or guar gum resulted in a decrease in the rate of flavor release, as shown by a reduced maximum intensity of the dynamic flavor release curve for some of the highly volatile compounds. Since thickened solutions of similar viscosity did not show the same flavor release, their results showed an influence of both viscosity and binding on the release of flavor. Although these studies have advanced our understanding of the effect of the food matrix on flavor release, there have been few data sets on flavor release subjected to interpretation using mathematical models.

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S0021-8561(97)00994-1 CCC: $15.00
Published on Web 06/03/1998

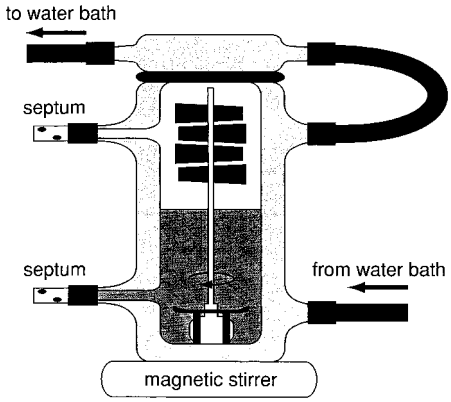

Figure 1. Schematic diagram of experimental setup to determine the release of diacetyl from a range of liquid gelatin concentrations and different stirring rates, at 37 °C.

The development of well-defined methods that can be described in terms of a mathematical model is imperative to develop our understanding of flavor release and perception. The data collected from such a method can then be interpreted using a mathematical model, which allows the development of further hypotheses to be tested. The aim of this paper is to present data from a well-defined experimental setup used to measure flavor release as a function of time from liquid gelatin solutions. We report two experiments: in one, the viscosity is increased with constant stirring, whereas in the other, the viscosity is kept constant but the stirring rate is varied. The data are also interpreted using mathematical models, providing a more complete interpretation of the physical factors affecting flavor release. From theoretical considerations one would expect that an increase in viscosity would to lead to a decrease in the mass transfer coefficient, whereas an increase in the stirring rate would lead to an increase in the mass transfer coefficient. Data are presented that validate a previously published mathematical model to describe flavor release from liquids containing aroma binding molecules (Harrison and Hills, 1997).

MATERIALS AND METHODS

Vessel. The experimental setup, including the design of the vessel, is shown in Figure 1. The 100 mL glass vessel has a low sidearm for the injection of the liquid sample and an arm in the headspace area for sampling of the air space. A round flea is connected to a vertical rod, which is connected to four paddles, which allows the airspace to be stirred at the same rate as the liquid, thus avoiding the buildup of any concentration gradient in both the liquid and the air. When the sample is inserted in the vessel, the liquid volume is 33 cm^3, the gas volume is 67 cm^3, and the surface area of the vessel is 12.6 cm^2 (vessel has a 4 cm diameter). The sidearms are closed with a Mininert screw-cap valve, with Teflon-coated seal, allowing sampling with a syringe and reducing the risk of leaks. The top is closed with a screw cap, lined with a Teflon seal. The sample is stirred with a magnetic stirrer (Heidolph, Germany), and a tachometer is used to select the number of rotations per second (rps) for the experiment. The setup is placed in an incubator to control the temperature at 37 °C.

Preparation of Gelatin Solution. Gelatin (food grade, bloom 78, provided by PB Gelatins U.K. Ltd, Mid Glamorgan) was dissolved in distilled water at 50 °C for 30 min using the Johnston-Banks indirect solution method (Johnston-Banks, 1990). All gelatin solutions are prepared the day before use, stored overnight at 4 °C, and warmed to 37 °C in a water bath (or incubator) (Jencons, Leighton Buzzard, U.K.).

Introduction of Flavored Solution in the Vessel. Half the gelatin solution (16.5 mL) was placed in the vessel, all kept at 37 °C. Double the concentration of diacetyl (100 mg/kg) was dissolved in the gelatin solution in a separate vial (volume = 44 mL) at 37 °C, which was kept sealed and as full as possible to avoid volatile losses. Once the diacetyl was fully dissolved, then 16.5 mL was injected via the bottom arm into the vessel, using a 25 mL syringe (Hamilton, Carnforth, U.K.), with a luer needle (51 mm length, 2.69 mm i.d., 10 gauge). The injection took 10 s and was done by injecting the sample near the bottom of the vessel, under the surface of the liquid gelatin already in the vessel, without the creation of turbulence. The injection was made with the top cap of the vessel slightly loosened, to allow the escape of air from the vessel, and it was tightened immediately after the injection was made. The syringe was not withdrawn from the vessel, to avoid possible leaking from the pierced septum. The magnetic stirrer and the stopwatch were started simultaneously, immediately after the flavored gelatin sample was introduced. To check the exact amount of gelatin solution introduced into the vessel, all vessels were weighed at various stages, but the reproducibility was shown to be good, with a coefficient of variation smaller than 1%, and no corrections for weight differences were necessary.

Headspace Analysis. Headspace sampling was done using a set of gastight 1 mL syringes (Hamilton), one syringe being used for each sampling point. Each syringe was filled with 500 μL of room air. The syringe was inserted into the top arm, the room air was injected to avoid a pressure change in the vessel due to sampling, and a sample of 500 μL of headspace was taken from the vessel. The syringe was capped with a septum and analyzed immediately after all samples in the sampling run had been taken. The first headspace sample was taken 15 s after introduction of the flavor. Samples were taken every 20 s during the first minute, every 30 s between 1 and 4 min, every minute between 5 and 8 min, and every 2 min thereafter. The run lasted 14 min. All samples were analyzed by gas chromatography (Hewlett-Packard 5890), using a splitless injection and isocratic analysis (60 °C). An HP-INNOWax capillary column was used (15 m × 0.53 i.d., 1 μm film thickness, from Hewlett-Packard, Palo Alto, CA) with helium carrier gas flow (10 mL/min), flame ionization detection, and injector and detector temperatures set at 170° and 300 °C, respectively. The retention time of diacetyl was 1.7 min. Daily calibrations in triplicate were done from 0.05 mg/kg diacetyl samples vaporized in air at 95 °C, by analyses of 500 μL of headspace, using exactly the same procedure as described for the headspace analysis. The typical coefficient of variation was 2% on a daily basis.

Experimental Designs. To determine the effect of gelatin concentration (0, 2.5, 5, 10, 15, and 20% w/v), the release of 50 mg/kg diacetyl into the headspace of the vessel was determined at 37 °C, using a constant stirring rate of 1.67 rps. To determine the effect of stirring rate (1.67, 2.50, 4.17, 5.50, and 7.33 rps) of a 15% w/v gelatin concentration, the release of 50 mg/kg diacetyl into the headspace of the vessel was determined at 37 °C. All experiments were run in triplicate. Equilibrium partition coefficients were determined in separate studies (at 37 °C) using brown screw-cap glass vials (40 mL), sealed with caps fitted with Mininert valves (Dynatech Precision Sampling Corp., Baton Rouge, LA).

Viscosity Measurements. The viscosity of the gelatin 78 bloom solutions was measured at 37 °C using a synchroelectric plate and cone viscometer (Wells Brookfield Micro, Stoughton, MA) with a 1.565° cone at shear rates from 23.0 to 115.0 s^{-1} (23.0 s^{-1} for gelatin concentrations 20 and 22.5% w/v, 46.0 s^{-1} for gelatin concentration 10% w/v, and 115.0 s^{-1} for gelatin concentrations between 0 and 16% w/v.

RESULTS AND DISCUSSION

One of the experimental requirements was introducing the flavored sample into the headspace, with minimal escape of the volatile into the headspace, such that at time 0 the concentration of volatile in the headspace would be near 0. This is an important constraint, since the release of flavor from a food in the

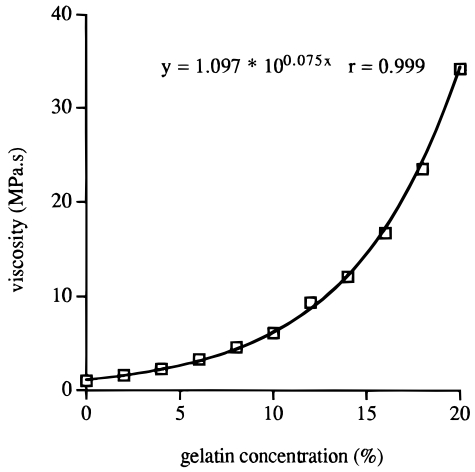

Figure 2. Viscosity (Mpa·s) of aqueous solutions containing increasing gelatin concentrations, determined at 37 °C. All measurements were done five times; average coefficient of variation was 8.2%.

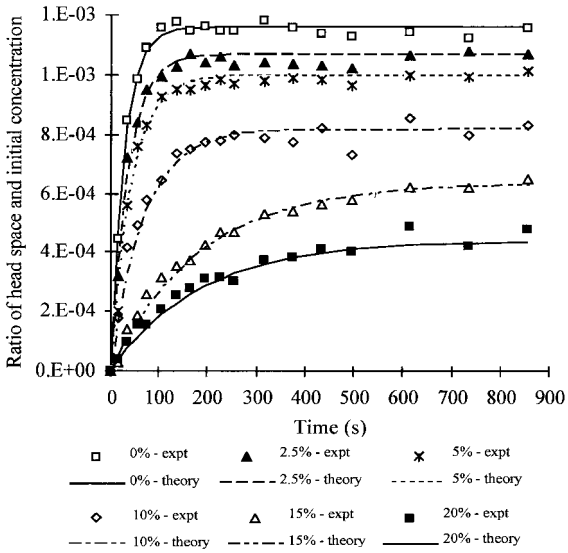

Figure 3. Diacetyl release from aqueous solutions containing 0, 2.5, 5, 10, 15, and 20% w/v gelatin at 37 °C. All measurements were done in triplicate; average coefficient of variation was 7.1%. The curves are fitted using the mathematical model.

mouth would similarly start under these conditions. People do not normally chew a mouthful of a food for more than a few minutes, hence the need for measurements of the early part of the release curve, that is, the release rates rather than just the equilibrium values, which may be much longer to attain. Although the release rates were expected to be fast, equilibrium partitioning conditions for some foods may take too long to establish during normal eating. Hence, it is probable that the initial release curve is important for flavor perception during eating, in addition to physiological and psychological aspects outside the scope of this work.

All release curves presented in this paper are determined for the vessel; that is, they are calculated for the surface area of the vessel (12.6 cm^2). Figure 2 shows the viscosity increasing exponentially with increasing gelatin concentrations in aqueous solutions. The viscosity of the liquid is expected to influence the diffusion of a small molecule in the liquid, since the Stokes–Einstein equation predicts that the diffusion is dependent on the square root of the viscosity. Although under the dynamic conditions used for the release measurements the bulk transfer of flavor is not expected to be diffusion controlled, the release is diffusion controlled at the interface and diffusion will influence the release rate by affecting the mass transfer rate, a theoretical consideration that is predicted to affect the release and discussed further below (Harrison et al., 1997; Harrison and Hills, 1997).

To mimic physical processes during eating and to allow interpretation of the resulting data, the experimental design of the procedure needed to be very precisely defined. Double the required concentration of diacetyl was mixed in half the gelatin and injected under the surface of half the gelatin without diacetyl already in the vessel. It was assumed that most of the diacetyl would remain under the surface of the unflavored gelatin already in the vessel, thus keeping the concentration of diacetyl in the headspace near 0 until the start of the experiment, when mixing was started immediately after the flavored gelatin introduction. It was further assumed that mixing of the gelatin solutions, both having the same gelatin concentration and being of the same temperature, would be fast. The procedure was tested by using a red dye in the gelatin solution injected into the vessel. Results showed that the colored gelatin rapidly distributed itself once stirring started at 1.67 rps, and within 10 s the entire solution appeared equally red.

Effect of Gelatin Concentration. The release of diacetyl as a function of time from aqueous solutions containing increasing concentrations of gelatin (0–20% w/v) is shown in Figure 3. To interpret the data from these results, a number of sections from the curves need to be considered. These included the initial slope of the curves, that is, the initial release rates, and the plateau values, that is, the equilibrium conditions and the time required to reach equilibrium. To interpret the data, a number of assumptions were made. First, the flavor concentration in the solution remained constant, since the losses into the headspace were small. Second, since all solutions and the airspaces were stirred at 1.67 rps, it was assumed that the flavor concentration remained well mixed within both the sample and the airspace and no concentration gradients were formed within the sample or the air.

The initial release rate of diacetyl from water under the test conditions was very rapid, as can be seen from the initial slopes of the linear part of the release curves (Figure 3). As the concentration of gelatin increased, the initial release rates of diacetyl decreased. The initial release rates are approximately linear, and the linear slopes for the first 1–2 min, which represent the initial release rates, were calculated and are shown in Table 1. The high r values indicate a good fit. These initial release rates appear to be linearly dependent on the gelatin concentration, as shown in Figure 4.

The diacetyl release from the aqueous concentration reached a plateau value after 2 min, as shown in Figure 3. The time required to reach this plateau increased with the concentration of gelatin, and the concentration of diacetyl released in the headspace above the 20% w/v gelatin solution appears to approximate its equilibrium after 1000 s. From an examination of the curves obtained for the 0–20% w/v gelatin concentrations, there appears to be a decrease in the equilibrium headspace concentration with increasing gelatin concentration, indicating an interaction between gelatin and diacetyl. Some form of binding between gelatin and

Table 1. Slopes and Intercepts Calculated from the Linear Part of the Release Curves of 50 mg/kg Diacetyl in Aqueous Solutions of Gelatin, All Stirred at 1.7 rps and Kept at 37 °C

gelatin concn (% w/v)	n^a	slope (ppm·s^{-1} × 10^{-3})	intercept (ppm × 10^{-3})	r	$K_{effga}{}^b$ (×10^{-3})	$K_{ga}{}^c$ (×10^{-3})
0	4	0.894	4.995	0.965	1.16	1.78
2.5	4	0.783	3.031	0.973	1.07	
5	4	0.715	0.279	0.994	1.00	0.874
10	4	0.458	1.528	0.977	0.82	0.507
15	6	0.160	0.149	0.998	0.64	0.313
20	7	0.090	0.967	0.981	0.44	0.250

[a] n equals the number of time points on the linear part of the slope, in triplicate. [b] K_{effga} is the effective equilibrium partition coefficient taken from the curve. [c] K_{ga} is the equilibrium partition coefficient, determined in separate experiments.

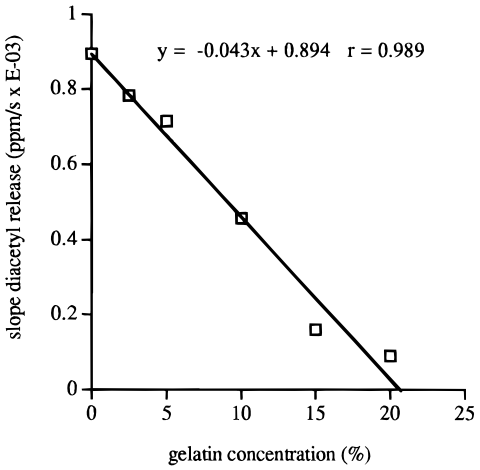

Figure 4. Rates of diacetyl release (ppm/s × 10^{-3}) as a function of the aqueous gelatin concentrations at 37 °C.

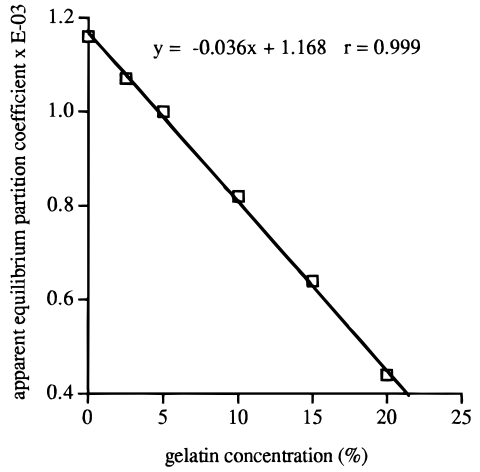

Figure 5. Effective equilibrium partition coefficients of diacetyl (K_{effga}) as a function of aqueous gelatin concentrations at 37 °C.

diacetyl is expected to reduce the free concentration of diacetyl and is reflected in a lower equilibrium partition coefficient, because the bound diacetyl is effectively not available to partake in the air/liquid equilibrium. In Table 1 the effective equilibrium partition coefficients taken from the curve, the K_{effga} values, are shown, and these K_{effga} values are plotted as a function of the gelatin concentration in Figure 5. The data fit excellently on a linear line (r = 0.999), showing that the effective equilibrium partition coefficients decrease linearly as the gelatin concentration increases.

Thus, there are two effects influencing the release of flavor. Binding of diacetyl to gelatin reduces the quantity of diacetyl available for release into the headspace, and this lower concentration of available diacetyl affects both the release rate, as expressed by the slope, and the equilibrium concentration, as expressed by the partition coefficient. Second, the increase in viscosity with increasing gelatin concentration will decrease the rate of diffusion of the diacetyl through the interfacial layer of the liquid. This will only affect the release rate and not the equilibrium concentration. The release rate of flavor will be dependent also on factors that are kept constant during this experiment, such as temperature and interfacial surface area, whereas the partition coefficient is expected to depend only on temperature.

A separate experiment was undertaken to determine the partition coefficients between diacetyl and gelatin solutions in glass vials (volume = 40 mL), minimizing binding with the vial. At 37 °C equilibrium partitioning appeared to be reached after 5 h of incubation of the solution in a shaking water bath. The results indicated that binding occurs between gelatin and diacetyl, as shown in the lower K_{ga} values with increasing gelatin concentration (Table 1). The discrepancy between the two data sets shows that indeed the effective equilibrium partitioning data extracted from Figure 3 were not the true equilibria, since the equilibrium partitioning took much longer to establish, and were lower for all solutions containing gelatin. This shows that the binding equilibrium takes quite some time to establish. For modeling the data, the effective partitioning equilibria data were used.

Modeling Gelatin Concentration Data. Further interpretation of the data can be accomplished by the use of mathematical models, although there are few models available that have been verified. In general, the transfer of flavor across a solid−liquid interface, such as the product−saliva interface, is reasonably well described by the two-layer stagnant film theory of interfacial mass transfer (Hills and Harrison, 1995; Harrison and Hills, 1996), whereas flavor release from liquids into air, such as from liquid foods into the oral cavity, seems best described by the penetration theory of interfacial mass transfer (Darling et al., 1986). Using penetration theory, Harrison et al. (1996) developed a mathematical model to describe flavor release from emulsions based on the assumption that the rate-limiting step is the resistance to mass transfer across the emulsion−gas interface. They assumed that partitioning of flavor molecules between the oil and aqueous phases is extremely rapid compared to the transport of flavor across the emulsion−gas interface. Their model suggests that the major physical factors affecting the rate of flavor release include the initial emulsion concentration, $c_e(0)$, the gas−emulsion partition coefficient, K_{ge}, and a viscosity-dependent interfacial mass transfer coefficient, h_D, to account for the effects of changing the oil fraction and droplet size. This model of Harrison et al. (1996) predicts that at low oil fractions h_D will increase because of the decreased shear viscosity, leading to faster flavor release from low-fat systems. Limited testing of the model confirmed its validity.

These authors also used penetration theory to develop a mathematical model to describe flavor release from liquid mixtures containing aroma-binding macromolecules (Harrison and Hills, 1997), such as the gelatin series presented here. Their model described the in-

gredient−aroma interaction as a major physical factor influencing both the release rate and the extent of flavor release from liquid foods. In stirred systems, such as those presented here, the bulk of the liquid will remain well mixed and have a constant concentration of the flavor volatile. Using penetration theory, the rate at which volatiles are released from a stirred liquid depends on two factors: the rate of diffusion, D, of a volatile in the liquid phase and the finite time, t_e, that liquid from the bulk phase is exposed to the gas phase as the system is agitated. Due to the impossibility of measuring the contact time, t_e, a parameter known as the mass transfer coefficient, h_D, is used (Coulson and Richardson, 1993), where

$$h_D = \sqrt{D/t_e} \tag{1}$$

According to penetration theory the rate at which flavor is released from the liquid, that is, h_D, is inversely proportional to the square root of viscosity and proportional to the square root of the stirring rate. Hence, an increase in the viscosity would cause a decrease in the rate of flavor release, while an increase in the stirring rate would cause an increase in the rate of flavor release. Two experiments are presented here: in one, the viscosity is increased with constant stirring, whereas in the other, the viscosity is kept constant but the stirring rate is varied. Thus, the release rate of diacetyl from the interfacial layer will be determined by diffusion and contact time, as expressed by the mass transfer coefficient.

The model also deals with binding of volatiles to the matrix and is based on two assumptions. The first assumption is that the aroma−polymer interaction is always at equilibrium and can be described by the reversible first-order reaction

$$\underset{c_{bf}}{\text{binder flavor complex}} \rightleftharpoons \underset{c_b}{\text{binder}} + \underset{c_{ff}}{\text{free flavor}}$$

where c_{bf}, c_b, and c_{ff} correspond to the concentrations of the bound flavor, binder, and free flavor in the aqueous phase, respectively. The total flavor in the solution is given by

$$c_{tf} = c_{bf} + c_{ff} \tag{2}$$

where c_{tf}, c_{bf}, and c_{ff} are the total, bound, and free flavor concentrations, respectively. The magnitude of the binding is described in terms of the binding coefficient, B (Overbosch et al., 1991) where

$$B = c_{bf}(t)/c_f(t) = K_b c_b(t) \tag{3}$$

where K_b is the binding constant and c_b is the concentration of binding polymer present in the aqueous phase. To test this theory using the release data for diacetyl from the different liquid gelatin solutions, first the binding constant, K_b, needs to be determined. This is done by fitting the effective partition coefficient, K_{effga}, of diacetyl between gas and the aqueous solution containing the binder using the following formula (Overbosch et al., 1991):

$$K_{effga} = K_{ga}/(1 + K_b c_b) \tag{4}$$

The data in Figure 5 have been fitted using a gas−aqueous partition coefficient, K_{ga}, of 1.20×10^{-3}. The best fit using this equation yielded a binding constant for diacetyl with gelatin, K_b, of 6 per percent gelatin. The binding constant, K_b, of 6 per percent gelatin was used to fit the theoretical curves on the time-dependent release data presented in Figure 3, using the mass transfer coefficient, h_D, as the fitting parameter for each gelatin concentration. Using these theoretical fits, a mass transfer coefficient value was obtained for each diacetyl release data from each gelatin concentration, ranging from 2.70×10^{-6} m/s for the diacetyl release from the aqueous solution to 0.40×10^{-6} m/s for the diacetyl release from the 20% w/v gelatin solution. The results are presented in Figure 6, which shows the mass transfer coefficient decreasing with increasing gelatin concentration.

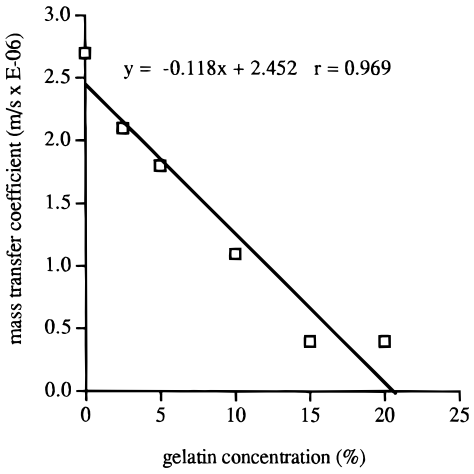

Figure 6. Mass transfer coefficient (m/s × 10^{-6}) of diacetyl as a function of gelatin concentration in aqueous solutions at 37 °C.

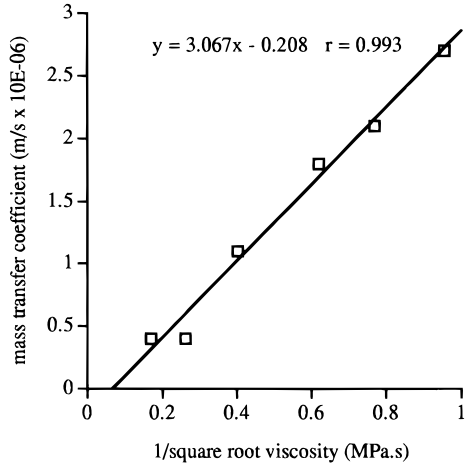

Figure 7. Mass transfer coefficient (m/s × 10^{-6}) of diacetyl as a function of 1/√viscosity of aqueous gelatin solutions at 37 °C.

An important correlation that should be investigated in the interpretation of these data is the relation between viscosity and the mass transfer coefficient. Theoretical considerations predict that the mass transfer coefficient is inversely proportional to the square root of viscosity of the gelatin solution. Figure 7 shows that 1/√viscosity plotted against the mass transfer coefficients shows an excellent linear correlation, with a very good linear fit. This correlation confirms the validity of the mathematical model to describe flavor release from solutions of increasing viscosity and suggests that the model is an adequate description of these

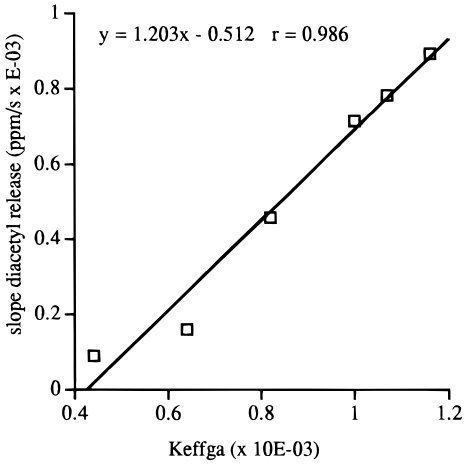

Figure 8. Rates of diacetyl release (ppm/s × 10^{-3}) as a function of the effective equilibrium partition coefficients of diacetyl (K_{effga}) in aqueous solutions containing 0, 2.5, 5, 10, 15, and 20% w/v gelatin at 37 °C.

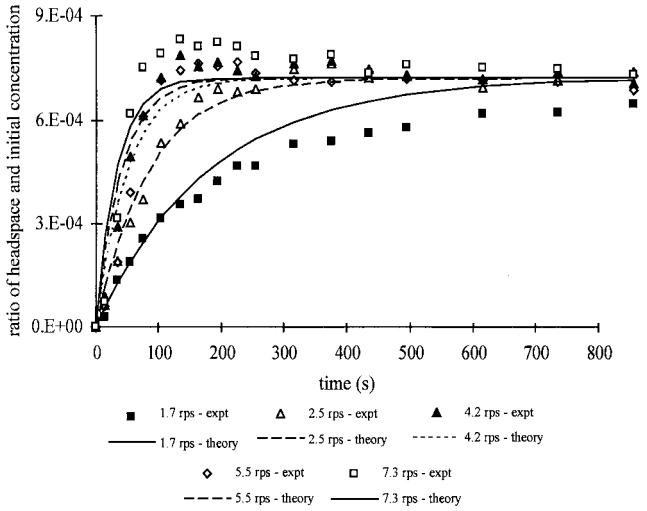

Figure 9. Effect of stirring rate (rps) on the release of diacetyl from an aqueous solution containing 15% w/v gelatin at 37 °C. All measurements were done five times; average coefficient of variation was 10.9%. The curves are fitted using the mathematical model.

experimental data. Hence, it may be possible to use viscosity measurements, which can be determined fairly rapidly, to derive intermediate mass transfer coefficients, which play an important role in the prediction of flavor release.

There is also a good correlation between the effective equilibrium partition coefficient and the release rate of diacetyl, as shown in Figure 8. However, the modeling implication is that the initial release rate is independent of effective equilibrium partition coefficient but depends on the amount of free flavor in the liquid phase (Harrison and Hills, 1997).

Effect of Stirring Rate. A second series of experiments on diacetyl release from liquid gelatin was performed to determine the effect of the stirring rate of a solution containing 15% w/v gelatin on the release of diacetyl. Figure 9 shows the time-dependent release curves of diacetyl from an aqueous solution containing 15% w/v gelatin and stirred at a stirring rate ranging from 1.67 to 7.33 rps. Since all solutions examined have the same composition, and only one parameter, the stirring rate, is varied, only the slopes of the initial release rates are expected to vary, whereas the equilibrium partitioning is expected to be the same for all of the curves. The initial release rate of diacetyl increased with an increase in the stirring rate, while the effective equilibrium partitioning coefficient after 15 min is the same for all stirring rates, except the most slowly stirred sample, which has not yet reached equilibrium value. The initial release rates are approximately linear, and the slopes for linear fits for the first 60−120 s, which represent the initial release rates, were calculated and are shown in Table 2. The high r values indicate a good fit.

Table 2. Slopes and Intercepts Calculated from the Linear Part of the Release Curves of 50 mg/kg Diacetyl an Aqueous Solutions of Gelatin (15%), Stirred from 1.7 to 7.3 rps and Kept at 37 °C

stirring rate (rps)	n^a	slope (ppm·s^{-1} × 10^{-3})	intercept (ppm × 10^{-3})	r
1.7	6	0.160	0.149	0.998
2.5	6	0.257	0.048	0.997
4.2	6	0.367	1.021	0.984
5.5	5	0.412	−2.351	0.986
7.3	5	0.541	−1.918	0.989

[a] n equals the number of time points on the linear part of the slope, in triplicate.

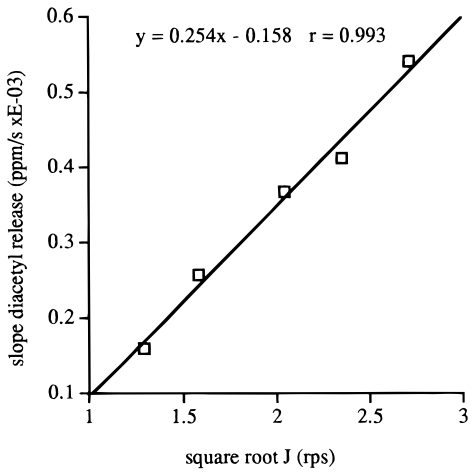

Figure 10. Rates of diacetyl release (ppm/s × 10^{-3}) as a function of the square root of the stirring rate (rpm) of an aqueous solution containing 15% w/v gelatin at 37 °C.

The release rate of diacetyl from an aqueous solution containing 15% w/v gelatin at 37 °C appears to increase linearly with the square root of the stirring rate, as shown in Figure 10. Coulson and Richardson(1993) suggested that release data should depend on the square root of the stirring rate. Stirring has been reported to increase the release of diacetyl from an aqueous solution, where stirring increased the release rate by approximately a factor 2 in comparison with the solution not being stirred (Salvador et al., 1994). These authors used a nonstirred sample, which would have allowed the possibility of a concentration gradient being formed in the liquid. By comparing different stirring rates in the data presented here, such a possible artifact was prevented.

Modeling Stirring Rate Data. As before, the experimental data have been fitted with the mathematical model specifically developed for these systems. As described above, the binding constant, K_b, of 6 per percent gelatin was used to fit the theoretical curves on the time-dependent release data presented in Figure 8, using the mass transfer coefficient, h_D, as the fitting parameter for stirring rate. Using these theoretical fits, a mass transfer coefficient value was obtained for each

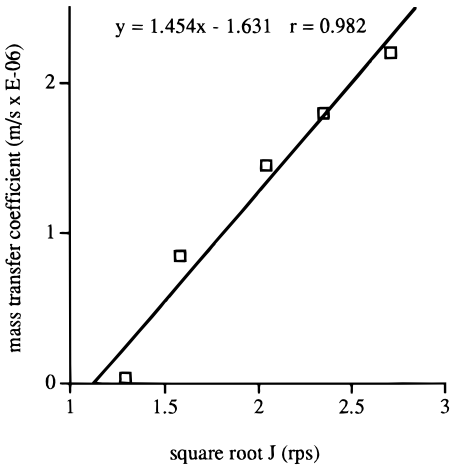

Figure 11. Mass transfer coefficient (m/s × 10^{-6}) of diacetyl as a function of 1/√stirring rate (rpm) of an aqueous gelatin solution containing 15% w/v gelatin at 37 °C.

stirring rate, ranging from 0.04 × 10^{-6} m/s for diacetyl release from the solution stirred at 1.67 rps to 2.20 × 10^{-6} m/s for diacetyl release from the 15% w/v gelatin solution stirred at 7.33 rps. The theoretical fits are less perfect than those for the concentration study. Imperfections in the experimental protocol may to some extent be to blame; for example, the modeling was applied by assuming a constant surface area. Higher stirring rates could have created a vortex, increasing the surface area with the stirring rate. To avoid the formation of a vortex, a high-viscosity solution (15% w/v gelatin) was chosen for this experiment. The data for the release curve obtained for the higher stirring rate indicate that the equilibrium concentration was reached rapidly in this experiment, possibly followed by a reabsorption of the diacetyl from the headspace into the liquid system. A possible explanation is imperfect mixing of the gelatin solution containing the flavor with the one containing no flavor, thus allowing more flavored concentration near the interface, resulting in a rapid release. Another possible explanation for this observation is that the equilibrium between free and bound diacetyl in the liquid was not obtained as quickly as was assumed for reversible binding but took some time to establish. This may indicate a chemical reaction between diacetyl and one or more of the amino acids of the gelatin molecule. Maier (1975) indicated that, for example, proline in a dry state was able to sorb high quantities of diacetyl, whereas arginine was able to sorb moderate quantities. Dumont and Land (1986) suggested that their data obtained in binding experiments with pea proteins indicated that diacetyl was bound and proposed that diacetyl may bind via direct interaction with arginyl residues in the protein. Since gelatin is known to have high concentrations of proline, binding interactions based on proline and arginine may occur in gelatin, analogous to the interpretation Dumont and Land (1986).

In Figure 11 the mass transfer coefficients are plotted as a function of the square root of the stirring rate. The results show that the mass transfer coefficient increases linearly with the square root of the stirring rate. This correlation confirms the penetration theory to describe interfacial transfer of volatile flavor molecules across the aqueous−gas interface.

In conclusion, the increase in both viscosity and stirring rate influenced the mass transfer coefficient. There was a linear increase in the mass transfer coefficient proportional to the square root of the stirring rate, whereas there was a linear decrease in the mass transfer coefficient inversely proportional to the square root of viscosity. These observations confirmed the validity of the mathematical model developed to describe these phenomena, derived from the penetration theory. The release rates over the early part of the time−concentration curves could also be shown to depend linearly on gelatin concentration and stirring rate, indicating that these may also be of use in predicting release rates.

ACKNOWLEDGMENT

We thank Kieran Black for viscosity measurements and partition coefficient determinations and Drs. Mark Springett and Wendy Brown for helpful discussions.

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Received for review November 24, 1997. Accepted April 15, 1998. This research was financially supported by a Competitive Strategic Grant from the Biotechnology and Biological Sciences Research Council.

JF970994L