Headspace and small-chamber studies of airborne diacetyl release from selected food flavoring mixtures: activity coefficients and air modeling implications

Paul K. Scotta*, Anders Abelmannb, Steve Hoytc and Brent D. Kergerd

aCardno ChemRisk, Pittsburgh, PA, USA; bCardno ChemRisk, Chicago, IL, USA; cEnvironmental Analytical Services, Inc., San Luis Obispo, CA, USA; dExponent, Inc., Irvine, CA, USA

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Laboratory studies were conducted to evaluate airborne release of diacetyl from selected mixtures simulating butter flavorings added to foods. The test materials included diacetyl (97% purity); 0.015%, 0.15%, 1.5%, and 3.0% diacetyl in a water/propylene glycol mixture; 1.5% diacetyl in deionized water or soybean oil; and 3% or 6% diacetyl in a commercial steam distillate from milk fermentation known as “butter starter distillate.” Diacetyl was quantified by gas chromatography with flame ionization detection. Expected concentration-dependent emission patterns based on liquid diacetyl content were demonstrated, but were significantly altered by mixture composition. Soybean oil and deionized water more readily released diacetyl when compared with starter distillate, propylene glycol solutions, and pure diacetyl. Measured diacetyl concentrations under static headspace and dynamic flow-chamber conditions were compared to estimated concentrations utilizing Raoult’s law with published and fitted activity coefficient corrections for each mixture, indicating that published coefficients often understated the measured concentrations. It is concluded that headspace (static) and small-chamber (dynamic) measurements of airborne diacetyl provide data to assist in validating model-estimated airborne diacetyl concentrations by using mixture-specific activity coefficients. Implications of these empirical data for validating exposure estimates for diacetyl based on near-field/far-field modeling in workplace settings are discussed.

Keywords: butter flavoring; occupational exposure assessment; human; UNIFAC activity coefficient; diacetyl; air dispersion modeling

Introduction

Diacetyl, also known as 2,3-butanedione and diacetyl, naturally occurs in a variety of consumer products including many beverages (e.g., tea, coffee, beer, wine, milk, and citrus juices) and foodstuffs (e.g., butter, yogurt, cheese, chicken and beef, and an array of fruits and vegetables) (Aghlara et al. 2009; Bartowsky and Henschke 2004; Burdock 2005; Gallardo-Escamilla, Kelly, and Delahunty 2005; NTP 2005). In addition, since the early 1900s, diacetyl has been manufactured in large quantities in the United States, and has been used in a variety of applications, including as a flavoring agent (food additive), a preservative agent in therapeutic drugs and food products, a reactant in many chemical and biochemical reactions, and a component of fragrances, dental composite materials, and hair bleaching compositions (NTP 2005; Opdyke 1979). As a food additive, diacetyl is used to convey a buttery or butterscotch flavor in many consumer products (Fujioka
Currently, diacetyl is included on the U.S. Food and Drug Administration's (FDA’s) Generally Recognized as Safe (GRAS) list for food additive uses (FDA 1983).

Recently, some concern has been expressed regarding workers who have allegedly developed severe respiratory disorders following exposure to diacetyl and other butter flavoring compounds (e.g., Kreiss 2013; Kreiss et al. 2002). Specifically, since the early 2000s, the National Institute for Occupational Safety and Health (NIOSH) has evaluated many facilities at which diacetyl or diacetyl-containing flavorings were used (e.g., microwave popcorn production, food flavoring production, etc.). NIOSH concluded based on their studies that occupational exposure to diacetyl may be associated with development of severe respiratory disease, including the rare condition bronchiolitis obliterans (BO), in highly exposed workers (NIOSH 2011). In response to these activities, diacetyl has largely been phased out from use in food flavorings; instead, 2,3-pentanedione and other diketones have been introduced as alternatives as they possess similar “butter-like” qualities (Anderson et al. 2013; Potera 2012; Boylstein 2012).

Due to these concerns over worker health and safety, various occupational exposure limits (OELs) for diacetyl have been proposed or recommended. In 2011, NIOSH published a draft criteria document regarding occupational exposure to diacetyl, in which a 15-minute Short-Term Exposure Limit (STEL) of 0.025 parts per million (ppm), and an 8-hour time-weighted average (8-hour TWA) Recommended Exposure Limit (REL) of 0.005 ppm were proposed (NIOSH 2011). Similarly, ACGIH adopted Threshold Limit Values (TLVs) for diacetyl in 2012, 0.02 ppm as a 15-minute STEL, and 0.01 ppm as an 8-hour TWA (ACGIH 2012). The NIOSH-recommended OELs have not been finalized to date, although they more recently presented an update to their quantitative risk assessment of diacetyl, which included an associated occupational limit of 0.06 ppm based on a human equivalent BMCL (benchmark concentrations lower confidence limit) of 1.4 ppm from bioassay data on male rat lung eosinophilic inflammation and use of a 24-fold uncertainty factor (NIOSH 2013). The European Commission (EC) (2013) published draft-recommended diacetyl OELs of 0.1 ppm as a 15-minute STEL and 0.020 ppm as an 8-hour TWA, which also have not been finalized, whereas the German MAK Commission has published a recommended 8-hour OEL for diacetyl of 0.02 ppm (Deutsche Forschungsge- meinschaft 2015). Similarly, the U.S. Occupational Safety and Health Administration (OSHA) has not promulgated OELs for diacetyl. In addition, others have proposed various OELs for diacetyl. In particular, Maier et al. (2010) evaluated the results of studies of diacetyl exposure on respiratory effects in mice, and used modeling to derive an 8-hour TWA OEL of 0.2 ppm for diacetyl. The authors concluded that their OEL was supported by the current epidemiology literature of diacetyl-exposed workers. Egilman, Schilling, and Menendez (2011) also proposed an 8-hour TWA OEL of 0.001 ppm, based on their evaluation of the available literature.

Although it is unclear whether diacetyl is a causative factor for BO cases identified in various occupational surveys (Finley, Galbraith, and Weill 2008; Galbraith and Weill 2009; Pierce et al. 2015), there is increased interest in estimating the diacetyl concentrations to which various workers in the flavoring industry may be currently or historically exposed. Airborne emissions modeling may be a useful tool to augment field measurements used to estimate the exposures to workers and others in a wide variety of situations from this common butter flavoring and to provide screening level estimates of exposure in the absence of field measurements. For this study, a series of laboratory studies of diacetyl emissions from various diacetyl mixtures common to the flavoring industry was performed to help refine emission modeling estimates for these mixtures.
Methods

Static headspace and dynamic flow-chamber study

To improve emissions estimates due to evaporation and better characterize the non-ideal-
ity of different diacetyl mixtures, a series of laboratory measurements of approximated
steady-state diacetyl headspace concentrations due to the evaporative emissions of eight
different solutions containing diacetyl were performed. In addition, a bench-scale
dynamic flow chamber was used to characterize steady-state chamber diacetyl air
concentrations over time for three different diacetyl concentrations in a solution of propylene
glycol and water.

To simulate the variety of flavoring mixtures that may be used by different manufac-
turers, the headspace evaluation included pure diacetyl and eight different diacetyl solu-
tions: starter distillate with 3.0% and 6.0% diacetyl; a mixture of deionized water with
1.5% diacetyl; a mixture of soybean oil with 1.5% diacetyl; and mixtures of 54% v/v prop-
ylene glycol in water with diacetyl concentrations of 0.015%, 0.15%, 1.5%, and 3.0%
diacetyl. Such starter distillate mixtures are typically used to make butter or cheese flavor-
ings, while the soybean oil mixture is representative of generic flavoring ingredients that
may be oil or fat based. The propylene glycol/water and water mixtures represent mixtures
that may be used to make butter flavorings for candy. The source of the diacetyl for each
of these mixtures was food-grade diacetyl (2,3-butanedione) with a purity of 97%
(#B85307-100 ml, Sigma-Aldrich, St. Louis, MO). The starter distillate used to create an
aqueous mixture typically containing 3%−5% diacetyl was a steam distillate fraction from
fermentation of cow’s milk which met the specifications set forth by the U.S. FDA (2015).

For the headspace measurements in each of the nine solutions evaluated, 1−2 ml of
the diacetyl solution was placed into a Tedlar bag with 1 μl of octane (internal standard)
and 1000 ml of ultra-high purity nitrogen. The headspace was sampled using a syringe
after a minimum equilibration time of 30 minutes at room temperature (72−73 °F). Air
samples from the syringes were analyzed by gas chromatography with flame ionization
detection (GC/FID) in accordance with the U.S. Environmental Protection Agency (EPA)
Method TO-3 (U.S. EPA 1984).

For the dynamic flow-chamber study, a one-gallon (3.8 l) chamber was equipped with an
inlet and outlet so that air flow could be regulated to produce four changes per hour (Supple-
mental Materials Figure S1). For the chamber measurements, three mixtures of 54% propyl-
ene glycol in water with diacetyl concentrations of 0.015%, 0.15%, and 1.5% were used. For
each of the three mixtures, a 100 ml aliquot of each solution was placed into the chamber.
After sealing the chamber and applying a calibrated input air flow rate, the well-mixed cham-
ber headspace was sampled using a syringe at varying time intervals between 30 and
60 minutes. Again, air samples from the syringes were analyzed using the U.S. EPA Method
TO-3. Two or three trials were run at approximately 30, 45, and 60 minutes for each mixture
in order to assess steady-state chamber headspace concentrations.

The dynamic flow-chamber studies were conducted using syringe air samples that
were analyzed using a cryotrap concentrator loop attached to the same GC/FID sys-
tem utilized for the direct injection method that was applied for all other data. It
was subsequently determined by quality control comparisons to the direct injection
method findings that the cryotrap concentrator loop at higher diacetyl concentrations
was associated with a high variability in calibration curve measurements exceeding
10,000 parts per million by volume (ppmv) in headspace gas. The data indicated
that the cryotrap was inefficient in capturing all diacetyl, leading to overstated diace-
tyl response factors (i.e., smaller peak area per ng of diacetyl) on the GC/FID.
However, the chamber study data for 1.5% diacetyl involved concentrations below 10,000 ppmv diacetyl and provided consistent analytical data on the same analytical system (GC/FID) that was used for both the cryotrap and direct injection methods. Therefore, a uniform response factor correction was applied to the chamber study data in order to calculate the corrected analytical results presented for the dynamic flow-chamber studies (See Supplemental Materials Appendix A).

In order to compare the airborne diacetyl concentration across mixture types and mixture concentrations for the static headspace and chamber measurements, the airborne diacetyl concentrations were normalized to the liquid diacetyl content by dividing the airborne concentration in ppm in air by the liquid diacetyl concentration in ppm. A one-way analysis of variance (ANOVA) and subsequent post-hoc Tukey multiple comparisons were used to compare the average normalized diacetyl concentrations by mixture type for the static headspace measurements and by diacetyl concentration for the propylene glycol/water mixtures for both the static headspace and chamber measurements. The Shapiro–Wilks test for a normal distribution was performed on the data by mixture type and by diacetyl concentration for the propylene glycol/water mixtures to verify that the normalized diacetyl concentrations had a normal distribution.

**Estimation of activity coefficients by mixture type and diacetyl content**

Diacetyl emissions and the associated diacetyl air concentrations were estimated in order to compare standard modeling assumptions with the empirical diacetyl measurements for each mixture that were evaluated in the headspace and chamber studies. Static headspace concentrations for each of the mixtures were estimated using Raoult’s law (Zumdahl 1986) and chamber concentrations at approximate equilibrium were estimated using the method presented in Smith (2001) for the evaporation of volatile chemicals.

For each of the diacetyl mixtures, Raoult’s law was used to estimate the expected (under ideal gas conditions) headspace equilibrium concentration of diacetyl (Keil 2009; Zumdahl 1986). The activity coefficient for each mixture was estimated using the UNIFAC program. UNIFAC is a semi-empirical system for the prediction of non-electrolyte activity in non-ideal mixtures. It uses the functional groups present on the molecules that make up the liquid mixture to calculate activity coefficients (Fredenslund, Jones, and Prausnitz 1975). For the 1.5% diacetyl in soybean oil mixture, the soybean oil was represented in the UNIFAC calculation by the mass-weighted average of the functional groups for the five triacylglycerols (tristearin, tripalmitin, triolein, trilinolein, and trilinolenin) that contribute 94.4% of the total mass for all components of soybean oil (Hammond et al. 2005). To evaluate the sensitivity of this calculation on the use of the weighted average, the activity coefficient was estimated assuming a mixture composition of 1.5% diacetyl plus each of the individual triacylglycerols. The result was less than 1% different (4.84 for the weighted average and 4.80 using the individual components). In addition, the activity coefficients for each mixture were also estimated for each sample using the measured diacetyl headspace concentrations and Raoult’s law.

For the dynamic flow-chamber experiment, the open surface model of Smith (2001) was used to estimate the air concentration within the chamber assuming four changes per hour and the dimensions of the chamber. The activity coefficients estimated using either the UNIFAC method or the fitted activity coefficients were applied for comparison to the mean of measured diacetyl chamber concentrations (See Supplemental Materials Appendix B).
Modeling applications for workplace diacetyl concentrations

Using the activity coefficients estimated from the measured headspace concentrations for each mixture, near-field diacetyl short-term 15-minute and 8-hour TWA air concentrations were estimated for an example scenario involving a worker who transfers flavoring into a mixing tank using a hose. This example scenario was selected because pouring flavoring into a mixing tank is a common task for adding flavoring to various food products, including candy, snack foods, coffee, and other flavor-coated beverages (Curwin, Deddens, and McKernan 2014; Martyny et al. 2008). For this scenario, the worker is assumed to transfer 100 gallons (380 l) of diacetyl-containing flavoring into a 1000 (3800) gallon mixing tank during the course of 30 minutes twice (i.e., two batches) in one 8-hour work shift. The total mass of diacetyl in the container headspace is assumed to be emitted to the air over the 30 minutes of emptying process, e.g., during the filling of drums (50 gallons each) of the final product.

Headspace diacetyl concentrations within the container were estimated using Raoult’s law (Zumdahl 1986) assuming the tank has 380 l of headspace (10% of 1000 gallon or 3800 l) using the method for estimating emissions during transfer operations proposed by Keil (2009). Four diacetyl mixtures were evaluated: butter starter distillate with 3.0% diacetyl; the deionized water mixture with 1.5% diacetyl; the soybean oil mixture with 1.5% diacetyl; and the propylene glycol/water mixture with 1.5% diacetyl (See Supplemental Materials Appendix C). These mixtures were selected to represent typical diacetyl mixtures that may be used at various food manufacturing facilities that used diacetyl-containing flavorings (Curwin, Deddens, and McKernan 2014).

The estimated average diacetyl emissions for each of the four mixtures were then used in a two-zone near-field/far-field indoor air dispersion model (Nicas 2009) to estimate the short-term 15-minute and 8-hour TWA air concentrations. The near-field air concentrations during the transfer of the flavoring into the mixing tank were estimated assuming the near field was a hemispheric region around the intake with a radius of 1 ft (0.30 m). The worker was assumed to be in the near field attaching/detaching the hose for 30 seconds each time flavoring was added to the tank. Air movement between the near field and far field was assumed to be due to a random air speed of 0.2 m/s, which is the typical random air movement associated with an industrial setting (Burton 2002). The entire operation was assumed to be contained in a room with a 50 × 50 ft area and a ceiling height of 25 ft (i.e., 15 × 15 × 8 m³), which are dimensions representative of a large-scale manufacturing facility. An air exchange rate of four changes per hour was used, which corresponds to the low end of the range reported by Burton (2002).

Results

Static headspace and dynamic flow-chamber measurements

Table 1 summarizes the analytical results for the tested mixtures based on the static headspace and dynamic chamber measurements. A summary of individual diacetyl measurements is provided in Supplemental Table S1. The arithmetic mean of measurements for each mixture was used for comparison of actual versus estimated headspace and chamber concentrations based on Raoult’s law with or without UNIFAC activity coefficient corrections. Table 1 also presents normalized values for airborne diacetyl concentrations expressed in ppmv airborne diacetyl per ppm diacetyl in the liquid mixture.

Static headspace concentrations varied by diacetyl mixture composition and by liquid diacetyl concentration for the same mixture (Table 1), consistent with Raoult’s law. As
Table 1. Summary of saturated headspace and chamber concentration data.\(^a\)

| Type       | Mixture                  | Diacetyl (%)\(^b\) | Number of samples | Minimum  | Maximum  | Arithmetic mean | Standard deviation | Arithmetic mean | Standard deviation |
|------------|--------------------------|--------------------|-------------------|----------|----------|-----------------|-------------------|-----------------|-------------------|
| Headspace  | Pure diacetyl            | 100                | 12                | 69,192   | 118,472  | 92,213          | 18,135            | 0.092           | 0.0180            |
| Headspace  | Starter distillate       | 3                  | 4                 | 3056     | 3186     | 3127            | 58                | 0.104           | 0.0019            |
| Headspace  | Starter distillate       | 6                  | 4                 | 7223     | 7958     | 7496            | 326               | 0.125           | 0.0054            |
| Headspace  | Water                    | 1.5                | 4                 | 2400     | 2668     | 2486            | 121               | 0.166           | 0.0082            |
| Headspace  | Soybean Oil              | 1.5                | 8                 | 3329     | 5960     | 4429            | 1112              | 0.295           | 0.0740            |
| Headspace  | Propylene glycol/water   | 0.015              | 4                 | 21       | 24       | 22              | 1                 | 0.146           | 0.0086            |
| Headspace  | Propylene glycol/water   | 0.15               | 4                 | 95       | 225      | 143             | 57                | 0.095           | 0.0380            |
| Headspace  | Propylene glycol/water   | 1.5                | 6                 | 856      | 1712     | 1336            | 321               | 0.089           | 0.0210            |
| Headspace  | Propylene glycol/water   | 3.0                | 4                 | 2516     | 2657     | 2591            | 73                | 0.086           | 0.0024            |
| Chamber    | Propylene glycol/water   | 0.015              | 2                 | 10       | 16       | 13              | –                 | 0.085           | –                 |
| Chamber    | Propylene glycol/water   | 0.15               | 8                 | 30       | 71       | 53              | 13                | 0.036           | 0.0083            |
| Chamber    | Propylene glycol/water   | 1.5                | 4                 | 475      | 658      | 545             | 83                | 0.036           | 0.0056            |

\(^a\) Data for each sample analyzed are presented in Supplemental Table 1.

\(^b\) Percent concentration of diacetyl in the liquid mixture used for the headspace or chamber analysis.

\(^c\) Measured air concentration of diacetyl in the headspace or the chamber.

\(^d\) The measured diacetyl air concentration divided by the diacetyl concentration in the mixture in units of ppm (Diacetyl Concentration)/(Percent Diacetyl \(\times 10,000\)).

\(^e\) The normalized diacetyl concentration for this mixture was significantly less \((p < 0.05)\) than the normalized concentration for the 1.5% soybean oil mixture.

\(^f\) The normalized diacetyl concentration for this mixture was significantly less \((p < 0.05)\) than the normalized concentration for the 1.5% water mixture.

\(^g\) The normalized diacetyl concentration for this mixture was significantly less \((p < 0.05)\) than the normalized concentration for the 0.015% propylene glycol/water mixture.

\(^h\) The normalized diacetyl chamber concentration for this mixture was significantly less \((p < 0.05)\) than the corresponding normalized diacetyl headspace concentration.

Insufficient samples \((n < 3)\) to calculate a standard deviation.
expected, the highest diacetyl headspace concentration was associated with pure diacetyl, which had an average headspace concentration of around 92,000 ppmv. In regards to the diacetyl mixtures tested, the highest absolute headspace concentration of diacetyl was observed for the 6% starter distillate mixture and the lowest was observed for 0.015% diacetyl in propylene glycol/water (Table 1). When all static headspace concentrations for tested mixtures were plotted versus liquid diacetyl concentration in the mixture, a direct linear relationship was identified (Figure 1, Panel A).

Airborne diacetyl concentrations that were normalized to the liquid diacetyl content were calculated in order to compare apparent mixture effects on static headspace diacetyl concentrations (Table 1; Figure 2, Panel A). The normalized data were tested to see if they had a normal distribution and the results are presented in Supplemental Table S2. Comparison of the normalized values in Table 1 using a one-way ANOVA and subsequent Tukey multiple comparison tests indicated that the soybean oil released the most diacetyl (0.295) when expressed as ppm in air per ppm in liquid; the normalized value for soybean oil was significantly greater ($p < 0.0001$) than all other group means (Figure 2, Panel A). The normalized value for 1.5% diacetyl in deionized water (0.166) was significantly greater than that for pure diacetyl and for 1.5% diacetyl in propylene glycol/water (Figure 2, Panel A). None of the other group means were significantly different from pure diacetyl when the normalized values were statistically compared ($p > 0.05$). The propylene glycol–water mixtures showed a higher normalized value at 0.015% diacetyl (0.146) that was significantly greater ($p = 0.0066$ to $p = 0.027$) than those for the higher diacetyl content propylene glycol–water mixtures (0.15%, 1.5%, and 3%) that were between 0.085 and 0.095 in a decreasing trend of mean values (Figure 2, Panel B).

The dynamic flow-chamber studies of diacetyl in propylene glycol–water revealed a similar strong correlation between airborne diacetyl and liquid content (Figure 1, Panel B) as seen in the static headspace studies. These chamber air measurements were considered to represent near steady-state conditions achieved after 60 minutes of chamber operation based on time studies showing a plateau in absolute diacetyl concentrations at 50–60 minutes for all tested mixtures (Supplemental Figure S2a and S2b). The dynamic flow chamber showed absolute diacetyl concentrations lower than the corresponding static headspace concentrations, with the static headspace concentration ranging from approximately twofold to threefold higher than the corresponding dynamic flow-chamber concentration (Table 1). When the normalized values were compared between the headspace and chamber measurements (Figure 2, Panel B), the chamber values at 0.15% and 1.5% diacetyl content were significantly lower than the static headspace values with corresponding diacetyl content. As seen with the static headspace testing of the propylene glycol–water mixture, the lowest liquid diacetyl content mixture (0.015%) had the highest normalized chamber concentration of diacetyl (0.086); however, this observation was based on only two measurements due to a lost sample (Table 1; Figure 2, Panel B).

**Activity coefficients and comparisons of measured and estimated diacetyl concentrations**

**Static headspace measurements**

Table 2 provides the calculated mixture-specific activity coefficients for static headspace diacetyl concentrations either predicted based on the UNIFAC method or fitted based on the ratio of current study mean measurements divided by Raoult’s law predicted values. The UNIFAC method activity coefficients by mixture were highest for deionized water,
followed by starter distillate, soybean oil, and propylene glycol–water. The fitted activity coefficients were generally higher than the UNIFAC method values except for soybean oil, where the fitted activity coefficient was 11-fold lower than the UNIFAC value. The fitted value for pure diacetyl was 1.23, higher than the UNIFAC value of 1.00 (i.e., based

Figure 1. Relationship between airborne and liquid diacetyl concentrations for diacetyl in a propylene glycol–water solution in static headspace (Panel A) and dynamic flow-chamber (Panel B) tests.
Figure 2. Comparison of measured headspace and chamber diacetyle concentrations normalized to liquid diacetyle content (ppm in air per ppm in liquid) for selected mixtures (Panel A) and for selected concentrations of diacetyle in a propylene glycol–water mixture (Panel B). Statistically significant differences in the normalized diacetyle content are presented in Table 1.
solely on Raoult’s law). Apparent outliers for the fitted activity coefficients included deionized water at 1.5% diacetyl content and propylene glycol–water at 0.015% diacetyl content, each being about twofold higher than the UNIFAC value (Table 2). The propylene glycol–water data for 0.15%, 1.5%, and 3% diacetyl showed a parallel trend of decreasing empirical activity coefficient with increasing diacetyl content, with the fitted values being 20%–34% higher than UNIFAC values. Confidence limits for the fitted activity coefficients generally included the UNIFAC value for pure diacetyl and for propylene glycol–water mixtures with 0.15%–3% diacetyl (Table 2).

Figure 3 (Panels A and B) compares the estimated static headspace diacetyl concentrations using the UNIFAC method activity coefficients to the average measured concentrations for each of the eight diacetyl mixtures tested. Panel A shows that UNIFAC activity coefficients tended to understate the measured diacetyl concentration for all aqueous mixtures, and significantly overstated the measured diacetyl concentration associated with the soybean oil vehicle (Figure 3). Panel B presents the predicted static headspace concentrations for four diacetyl mixture concentrations in the propylene glycol/water mixture, again indicating that UNIFAC activity coefficients tended to understate the measured diacetyl concentrations (Figure 3). The differential was twofold at the lowest diacetyl concentration of 0.015%, but increasingly closer prediction was observed with increasing liquid diacetyl concentration. The relative percent differences between the UNIFAC estimates and the measured values [(Estimated – Measured)/Measured] were 35%, 25%, and 18% at 0.15%, 1.5%, and 3% diacetyl, respectively. The relationship between empirically based diacetyl activity coefficients in the current study and the UNIFAC-based predictions is illustrated in Supplemental Materials Figure S3, indicating that aqueous-based solutions may be understated when using the UNIFAC method, while soybean oil solutions are apparently overstated by the same method.

Table 2. Activity coefficients estimated using UNIFAC method and by fitting to saturated headspace air concentrations.

| Mixture                  | Diacetyl (%)a | UNIFACb | Fitted to headspace concentrationsc |
|-------------------------|---------------|---------|-------------------------------------|
| Pure diacetyl           | 100           | 1.00    | 1.24 ± 0.24 (12)                    |
| Starter distillate      | 3             | 5.03    | 6.47 ± 0.12 (4)                     |
| Starter distillate      | 6             | 4.94    | 7.77 ± 0.34 (4)                     |
| Water                   | 1.5           | 5.08    | 10.5 ± 0.52 (4)                     |
| Soybean oil             | 1.5           | 4.84    | 0.44 ± 0.11 (8)                     |
| Propylene glycol/water  | 0.015         | 2.65    | 5.48 ± 0.32 (4)                     |
| Propylene glycol/water  | 0.15          | 2.65d   | 3.57 ± 1.43 (4)                     |
| Propylene glycol/water  | 1.5           | 2.62d   | 3.27 ± 0.78 (6)                     |
| Propylene glycol/water  | 3.0           | 2.60    | 3.10 ± 0.087 (4)                    |

aPercent concentration of diacetyl in the liquid mixture used for the headspace analysis.
bActivity coefficient estimated using the UNIFAC methodology based on the mixture components.
cActivity coefficients estimated by fitting the estimated headspace concentration to the measured headspace concentration for each sample. The arithmetic mean ± standard deviation (N) is presented based on the data in Supplemental Table 1.
dThe UNIFAC estimate for activity coefficient is within the 95% confidence interval of the activity coefficients estimated by fitting the activity coefficient using the measured headspace concentrations.
Figure 3. Comparison of measured and UNIFAC estimated airborne diacetyl concentrations for selected mixtures (Panel A) and for selected concentrations of diacetyl in a propylene glycol–water mixture (Panel B). Error bars represent the standard deviation.
Dynamic flow-chamber measurements

Figure 4 presents airborne diacetyl concentrations in the dynamic flow chamber, either measured or estimated using the UNIFAC estimated and fitted activity coefficients for static headspace testing of the propylene glycol–water mixtures. For the 1.5% diacetyl solution, the UNIFAC and empirically fitted activity coefficients resulted in predicted diacetyl concentrations that were 10% and 13% of the average measured value, respectively (Figure 4). At 0.15% diacetyl content the UNIFAC and empirically fitted activity coefficients led to predicted diacetyl concentrations that had relative differences of 38% and 83%, respectively, whereas at 0.015% diacetyl content, the corresponding relative differences between the measured and predicted concentrations were 46% and 15% (Figure 4). In two of three cases (0.15% and 1.5% diacetyl), the UNIFAC activity coefficients tended to produce results that were closer to the measured chamber concentrations when compared to the fitted activity coefficients which were uniformly higher than the measured values (Figure 4). However, the UNIFAC coefficients considerably understated (by twofold) airborne diacetyl concentrations for emissions from the mixture with 0.015% diacetyl content.

Application to workplace exposure modeling

Figure 5 presents the estimated short-term 15-minute (Panel A) and 8-hour TWA diacetyl air concentrations (Panel B) associated with emissions from four flavoring mixtures for the example workplace exposure scenario. The emission rates for each of the four mixtures for this example scenario were estimated using either empirically fitted activity
Figure 5. Comparison of workplace diacetyl exposures using UNIFAC-based and static headspace fitted activity coefficients for selected flavoring mixtures: 15-minute short-term (Panel A) and 8-hr time-weighted average (Panel B) airborne diacetyl concentrations.
coefficients based on the measured headspace data or the UNIFAC method activity coefficient shown in Table 2. The propylene glycol/water mixture with 1.5% diacetyl showed the lowest emission rate and the soybean oil mixture with 1.5% diacetyl showed the highest emission rate. The short-term 15-minute near field diacetyl air concentrations based on empirically fit activity coefficients ranged from 4.9 ppm for the propylene glycol/water mixture at 1.5% diacetyl to 16 ppm for the soybean oil mixture at 1.5% diacetyl; UNIFAC method values for the same mixtures ranged from 3.9 ppm to 177 ppm, respectively (Figure 5, Panel A). The 8-hour TWA estimates for the four mixtures using the empirically fitted activity coefficients ranged from 0.16 ppm for the starter distillate with 3% diacetyl to 0.43 ppm for the soybean oil mixture; UNIFAC method values for the same four mixtures ranged from 0.13 ppm (propylene glycol/water mixture) to 4.0 ppm (soybean oil mixture).

Discussion

This is the first study to examine the influence of mixture composition and liquid diacetyl content on the release of diacetyl from flavoring mixtures in both static headspace and dynamic flow-chamber studies. While liquid diacetyl concentrations were well correlated with airborne diacetyl concentrations under static or dynamic flow conditions (Figure 1), the data clearly demonstrate significant impacts of mixture type on release of diacetyl from liquid to airborne phases (Table 1; Figures 2 and 3). The data indicate that soybean oil releases diacetyl from the liquid phase to a significantly greater degree than do aqueous solutions. Among the aqueous solutions tested, diacetyl was released more readily from deionized water when compared to either butter starter distillate (steam distillate of milk fermentation containing diacetyl) used in dairy product flavorings or to a propylene glycol–water mixture utilized in candy flavoring (Table 1). With increasing liquid diacetyl concentrations in propylene glycol–water solution, a reduction in the normalized rate of diacetyl release (ppmv in air per ppm in liquid) was observed consistent with a “like dissolves like” attraction keeping diacetyl in solution, similar to observations on benzene recently reported by others (Hollins et al. 2013; Richter et al. 2013).

The UNIFAC method provides a method to calculate mixture-specific activity coefficients for adjusting from ideal gas conditions (Raoult’s law) to the likely diacetyl emissions from any given mixture of known composition. Our static headspace tests for pure diacetyl and selected mixtures suggest that the UNIFAC method slightly underestates airborne diacetyl concentrations for aqueous mixtures and appears to greatly overstate airborne diacetyl concentrations above soybean oil mixtures (Figure 3, Panels A and B; Table 2). However, soybean oil did release diacetyl at a significantly greater rate than pure diacetyl or any aqueous mixture when normalized to liquid diacetyl concentration (Figure 2, Panel A; Table 1). The UNIFAC assumptions utilized are clearly not predictive of diacetyl release from the soybean oil mixture utilized, and may reflect rapid redistribution to the oil surface due to the polarity of diacetyl under the conditions used in this study. This greater release of diacetyl from soybean oil may be an important factor in understanding past workplace exposures to diacetyl and other butter flavoring components in the microwave popcorn facilities where clustered cases of BO have been reported [reviewed by Kreiss (2012) and Kreiss (2013)]. Moreover, additional data and improved understanding of activity coefficients for diacetyl in aqueous solutions is important for accurately characterizing past workplace exposures, such as those occurring from the use of butter starter distillate in dairy product flavorings and the use of propylene glycol–water solutions in certain candies that include butter flavoring.
The impact of using UNIFAC-based and empirical headspace data for diacetyl activity coefficients on prediction of airborne diacetyl concentrations was examined in a dynamic flow chamber using liquid diacetyl content of 0.015%–1.5% in propylene glycol/water solutions. The data suggest that at 0.15%–1.5% diacetyl content the measured airborne diacetyl concentrations within the chamber were generally lower than both UNIFAC-based and empirically based predictions (Figure 4). Therefore, airborne diacetyl concentrations tended to be understated when applying either UNIFAC-based or empirically based activity coefficients for aqueous diacetyl mixtures with >0.1% diacetyl content. However, both the headspace and chamber studies revealed that airborne diacetyl concentrations normalized to the liquid diacetyl content were considerably higher in propylene glycol/water solutions with 0.015% diacetyl content (Figure 2, Panel A and B; Table 1). Based on the limited measurements available for the propylene/glycol solution with 0.015% diacetyl (n = 2), the UNIFAC method led to understated airborne diacetyl concentrations at 0.015% diacetyl content, while the empirically fitted values slightly overstated the measured values (Figure 4). These findings suggest that the weaker “like dissolves like” attractions in more dilute aqueous solutions of diacetyl may require empirically derived activity coefficients to assure that model-predicted airborne exposures are not understated.

The effect of applying either UNIFAC method-based or empirically based activity coefficients on model-predicted airborne diacetyl concentrations for selected mixtures was also examined. The scenario involved near-field worker exposures during the addition of diacetyl in four different mixtures to a tank of food product that is subsequently blended and packaged to make two food product batches per work day. The illustrations in Figure 5 indicate that 15-minute and 8-hour TWA concentrations based on the empirically fitted activity coefficients may vary up to nearly threefold depending upon vehicle in which the diacetyl flavoring was added. In addition, it is important to consider that activity coefficients for diacetyl will vary depending on the diacetyl content and the specific composition of the added flavoring mixture and of the final food product being studied. Many other site-specific conditions may need to be considered in order to assure accuracy, such as accounting for product and air temperatures, ventilation conditions, time-activity patterns of the exposed worker, presence of other flavoring chemicals in the flavoring mixture and/or the final food product, and other factors.

There are several limitations to the studies presented here. First, this was only a sampling of selected diacetyl flavoring mixtures. The current studies should be confirmed with more robust and rigorous testing of mixture-related and diacetyl content-related effects on airborne diacetyl concentrations under static and dynamic test conditions. Second, while the direct injection method for diacetyl utilized in this study avoids sampling errors due to losses related to humidity and/or inconsistent recovery from sampling media, the analytical variability was greater at higher diacetyl concentrations (e.g., for headspace tests of pure diacetyl) and this should be taken into account when interpreting the data. Third, small-chamber studies can be subject to greater variability associated with incomplete mixing and the effect of such variability was not rigorously evaluated in this study. Finally, the workplace modeling of near-field and far-field diacetyl concentrations in the current study is a relatively generic example that does not explicitly represent any particular workplace or product manufacturing setting. Rigorous assessment and inclusion of site-specific modeling parameters and the collection of workplace validation sampling data are recommended in order to assure that the outcome can be considered reasonably accurate. Therefore, it is difficult to directly compare these modeled estimates to any OELs, such as those currently proposed or recommended.
Conclusions
The data generated in this study indicate that diacetyl release from various mixtures varies
depending on the liquid diacetyl content as well as the mixture composition, and that these
factors should be taken into account when evaluating potential airborne emissions and
exposures to diacetyl. Soybean oil released diacetyl more readily than did aqueous solu-
tions when normalized to liquid diacetyl content, which may be important in understanding
differences in workplace exposures where these different forms of butter flavoring may be
utilized. Increasing concentrations of diacetyl in aqueous media can actually lead to
reduced release of diacetyl when considering liquid concentration-normalized values, prob-
ably due to “like dissolves like” attractive forces that keep diacetyl in solution. These
observations may be important for quantitative modeling of diacetyl emissions from flavor-
ing mixtures or food products, and in more accurately assessing activity coefficients for
any given mixture containing diacetyl. Accordingly, the collection of mixture-specific
empirical data and actual workplace airborne concentration data can be important for
model validation of past exposures to diacetyl and other volatile flavoring compounds.

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