Phenotypic differences and genetic parameters for methane concentration in BeefxDairy crossbred slaughter calves

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Abstract
Genetic selection for reduced methane emission and BeefxDairy crossbreeding are strategies to lower the climate impact of beef. Combined they require knowledge of phenotypes on methane production for BeefxDairy offspring. Slaughter calves sired by Belgian Blue, Charolaise and Angus sires to Holstein dams were compared to purebred Holstein calves. Heritability for methane concentration (CH4c) were estimated for Belgian blue crosses. Raw phenotypic means showed that CH4c was lower for calves born after Charolaise and Angus sires compared to Belgian Blue. The heritability of log transformed CH4c for Belgian blue crosses was estimated to be 0.44 (±0.08), and for log transformed CH4c adjusted for background CH4 it was 0.33 (±0.06). This is in line with heritabilities found for log transformed CH4c in Holstein cows, and will be the basis for further analysis in genetic architecture of methane traits in crossbred slaughter calves.

Introduction
Reducing methane production from the dairy cattle population through genetic selection is seen as one of the ways to reduce the carbon footprint of cattle production. An increasing practice for Danish dairy farmers is insemination of low genetic merit dairy cows with beef semen to increase the value of the slaughter calf and overall income of the farm (Ettema et al. 2017). This reduces carbon footprints per kg carcass compared with purebred dairy calves (Mogensen et al., 2015). The beef sires are currently selected on calving ease, youngstock survival and carcass traits (Davis et al., 2020), but new traits could possibly lead to higher efficiency and lower climate impact. Currently there is a lack of knowledge, whether the sire breed affects methane (CH4) emission of BeefxDairy offspring, but also if there is variation between sires. Methane concentrations measured relative to CO2 concentrations in the breath using the sniffer method has a high throughput and has been shown to be heritable in dairy cows (Lassen & Løvendahl, 2016). The aim of this paper is to investigate the distribution of CH4 concentration in different crosses and estimate the heritability of CH4 concentration in Belgian Blue (BBL) and Holstein (HOL) crossbred calves.

Materials & Methods
Data were collected in the period June 2020 until November 2021. Calves were born on dairy farms in Denmark between July 2019 and June 2021. Six slaughter calf producers purchased these calves at the age of 3-8 weeks, and in these herds the animals were later phenotyped. 2,224 calves were crossbred, either a BBL (n=1,689), Charolaise (CHA) (n=306) or Angus (ANG) bull (n=229) on a Holstein dam. Purebred HOL bull calves (n=429) were also phenotyped, to match the capacity of the system. Calves were raised on a pelleted concentrate ration and offered ad lib straw, in five herds. One herd fed total mixed ratio (TMR). Feed intake on pelleted feed and TMR was measured in feed boxes, the feed was weighed as the
calf entered and left the feed box. The calf entered the pens between 6 and 10 months of age and stayed in the pen for at least three weeks.

CO2 and CH4 concentrations were measured in feed boxes using the non-invasive infrared gas sensor (Guardian® NG, Edinburgh Instruments Ltd.). Three feed boxes were available for each pen and neighbouring pens shared a Guardian Sensor. The intake air tube for gas measurements were located in front of the calf when it was eating the feed. Feed intake was registered at every visit, whereas gas concentrations was measured in one of the six feed boxes connected to the Guardian Sensor. During visits with gas recording, CO2 and CH4 concentrations were registered every other second. Gas concentrations were adjusted for background concentrations and detection of eructations was made using double exponential smoothing (Difford et.al. 2016). Gas visits where required to contain at least one eructation with methane concentration >500 ppm and be of length longer than 50 seconds. The average CH4 concentration (ppm) (CH4c) of the visit was used for estimating genetic parameters. Twice daily the gas sensors were running a reference measurement to measure background gas concentration, which was used to adjust the methane concentration. The adjusted phenotype (CH4a) was calculated for all visits using the most recent reference measure. To ensure the residuals were normally distributed, both variables were transformed by the natural logarithm (logCH4c, logCH4a), to ensure normal distribution for the genetic parameter analysis.

Genetic analysis.
Due to sample size, only phenotypes from BBLxHOL calves were used for estimating genetic parameters. For the variance component estimation, using the average information restricted maximum likelihood algorithm (AI-REML), the statistical software DMU was used (Madsen and Jensen, 2008). The univariate model for estimating genetic parameters for logCH4c and logCH4a was the following:

\[
y = Xb + Z_1a + Z_2pe + e
\]

where \( y \) is the vector of phenotypic observations, \( b \) is the vector of fixed effects (herd-sensor age in weeks, date of the visit, month-year of birth, sex, feed ration, hour of the day), \( a \) is the vector of additive genetic effects, \( pe \) is the vector of permanent random effects and \( e \) is the vector of random residual effect. The incidence matrix, \( X \), assigns observations to fixed effects, \( Z_1 \) assigns the observations to random genetic effects and \( Z_2 \) assigns the observations to the permanent environmental effect. The distribution of the random effects was: \( \text{var}(a) = A\sigma_a^2 \), where \( A \) is the additive genetic relationship matrix and \( \sigma_a^2 \) is the additive genetic variance; \( \text{var}(pe) = I\sigma_{pe}^2 \) where \( I \) is an identity matrix and \( \sigma_{pe}^2 \) is the permanent environmental variance; \( \text{var}(e) = I\sigma_e^2 \) where \( \sigma_e^2 \) is the residual variance. Standard error of the estimates was estimated by calculating asymptotic standard errors. The repeatability was calculated as ratio between the sum of additive variance and permanent effect variance, and the phenotypic variance. The heritability was calculated as the ratio between additive variance and phenotypic variance.

Results
The logCH4 concentration for each cross and herd is shown in Figure 1. Descriptive statistics of visits and methane concentration are presented in Table 1. The number of visits varied between breed groups. The slaughter calf producer contributed with different number of calves due to difference in capacity at sights (farm1 = 288, farm2 = 1,583, farm3 = 62, farm4 = 71, farm5 = 554, farm6 = 105). The calves represented both sexes (bull=1,620, heifer=1,043). Phenotypic means and standard error of the mean of methane concentration are presented in Table 1.
Table 1. Phenotypic means and standard error of the mean (SEM) for methane visits and traits\(^1\) for each breed group\(^2\) of slaughter calves.

<table>
<thead>
<tr>
<th>Trait</th>
<th>BBLxHOL</th>
<th>CHAxHOL</th>
<th>ANGxHOL</th>
<th>HOL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Visits per animal (n)</td>
<td>45.4 (1.35)</td>
<td>21.0 (1.89)</td>
<td>23.8 (1.62)</td>
<td>40.0 (2.55)</td>
</tr>
<tr>
<td>Visit time (s)</td>
<td>43.8 (0.71)</td>
<td>42.5 (1.55)</td>
<td>49.1 (2.00)</td>
<td>39.7 (1.32)</td>
</tr>
<tr>
<td>CH4c (ppm)</td>
<td>1,404 (20.0)</td>
<td>1,066 (41.2)</td>
<td>1,091 (39.3)</td>
<td>1,361 (40.0)</td>
</tr>
<tr>
<td>CH4a (ppm)</td>
<td>1,029 (17.8)</td>
<td>729 (33.9)</td>
<td>768 (33.2)</td>
<td>1,076 (35.2)</td>
</tr>
<tr>
<td>logCH4c (ln(ppm))</td>
<td>7.09 (0.01)</td>
<td>6.74 (0.04)</td>
<td>6.85 (0.04)</td>
<td>7.05 (0.03)</td>
</tr>
<tr>
<td>logCH4a (ln(ppm))</td>
<td>6.73 (0.02)</td>
<td>6.34 (0.04)</td>
<td>6.47 (0.04)</td>
<td>6.79 (0.03)</td>
</tr>
</tbody>
</table>

\(^1\) CH4c = Methane concentration, CH4adj = Methane concentration adjusted for background, logCH4c = ln transformed methane concentration, logCH4a = ln transformed methane concentration adjusted for background  
\(^2\) BBLxHOL = Belgian Blue cross, CHAxHOL = Charolaise cross, ANGxHOL = Angus cross, HOL = Holstein

The genetic parameters were estimated on the 1,689 BBLxHOL calves. The results from the parameter estimation are shown in Table 2. The heritability of the traits are moderate for logCH4c and logCH4a, and all significantly different from zero. Repeatability for logCH4c and logCH4a is equal to the heritability.

Table 2. Estimated repeatability and heritability with asymptotic standard error for methane concentration traits\(^1\)

<table>
<thead>
<tr>
<th>Trait</th>
<th>R</th>
<th>h(^2) (SE)</th>
</tr>
</thead>
<tbody>
<tr>
<td>logCH4c</td>
<td>0.44</td>
<td>0.44 (0.08)</td>
</tr>
<tr>
<td>logCH4a</td>
<td>0.30</td>
<td>0.30 (0.06)</td>
</tr>
</tbody>
</table>

\(^1\) logCH4c = ln transformed methane concentration, logCH4a = ln transformed methane concentration adjusted for background

Discussion

This work is the first step on the road to achieving a methane phenotype for BeefxDairy crossbred calves. The level of CH\(_4\) concentration is higher than what was measured in Danish
Holstein cows, when methane was measured in the milking robot (Difford et al., 2020; Manzanilla-Pech et al., 2021). Mean CH₄ concentration CHAxHOL and ANGxHOL was lower compared to BBLxHOL and purebred HOL (Table 1). Variation within groups is large (Figure 1), so conclusions should be made with caution. No difference in CH₄ was seen between herds, and variation was not consistent across herds. This could be due to the use of TMR in one case (Herd 4).

The repeatability was equal to the estimated heritability. This is caused by the estimated permanent environmental effect being zero. This conflicts with other studies in dairy cows, where the repeatability was a lot higher than the heritability (Difford et al., 2020). The standard error in the estimates in this study is however large. Increasing the sample size will provide a lower estimate of the standard error and a higher accuracy.

The heritabilities for CH₄ concentration was estimated with a large standard error, but is in range with CH₄ traits from other studies (Difford et al., 2020; Hayes et al., 2016; Lassen & Lovendahl, 2016, Manzanilla-Pech, 2021). Log transformed CH₄ concentration is used when estimating parameters on CH₄ concentration to meet the requirement of normal distributed residuals (Difford et al., 2016). Methane concentration can be extrapolated to a daily methane production which has a moderate heritability in lactating HOL cows (Manzanilla-Pech, et al. 2021). CHAxHOL and ANGxHOL had a lower phenotypic mean CH₄ concentration compared to BBLxHOL and purebred HOL. The heritability of CH₄ concentration was estimated to be moderate in BBLxHOL crosses. Further work will develop this phenotype further and investigate the genetic architecture of the trait.

References