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Abstract

# Assessing the usefulness of fat content and milk yield data gained during ICAR farm tests of milk recording and sampling devices to estimate carry-over in milking systems

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Carry-over between milkings can affect sampling results that are used for herd management, breeding or diagnostic purposes in dairy cows. Giving an estimate of a milking system's carry-over therefore would be a useful additional part of the ICAR certification of a recording and sampling device.

Current methods of estimating or calculating carry-over between subsequent milkings require additional expenses for e.g. tracer chemicals or constituents of milk that can be used as a tracer and their analysis, respectively. Another factor required for these methods is time, which also translates into additional expenses. However, during an ICAR farm test of a recording and sampling device all relevant data required for an estimation of carry-over can potentially be recorded. This aim of this article is to evaluate the usefulness of these data (milk yields, fat contents, milking sequences) for estimating a given milking system's carry-over.

Data gained during ICAR farm tests are used in different statistical models (regression analysis, linear mixed model analysis with repeated measures) to estimate the carryover of the combination of milk recording device and sampling device under test. This includes at least four different manufacturers and different milking systems (three automatic milking systems, one conventional milking system).

Data from experimental farms are used as a base to create "virtual" herds of dairy cows. These herds are used to simulate different setups of an ICAR farm test: conventional milking systems and automatic milking systems with short and long sequences of subsequent milkings per day and milking time, as well as different levels of carry-over. Carry-over is then estimated in simulations using the above-mentioned statistical methods. The simulations indicate the level of carry-over that could be detected in ICAR farm tests as well as the inherent test power for the different setups.

The results of the simulations are compared to the ICAR farm tests with a similar setup, and as a conclusion possible options for future farm tests are derived.

Milk samples are used for several types of laboratory diagnostics, spanning from e.g. fat and protein content to somatic cell count and pregnancy or disease detection. For some of these lab diagnostics it is important to have an idea about the carry-over of the milking system the samples were gained from, since the robustness of the diagnostic

#### Introduction

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methods against carry-over may not always been given, and hence carry-over can affect the results. If the expected amount of carry-over is known, this knowledge can be used to assess the usefulness of a given sample. Lab methods can be adjusted to provide reliable results, or the sampling procedure in a given milking system can be adjusted to take samples in a way that reduces or avoids carry-over, if samples are taken for a specific purpose. Simply put, knowing a milking system's carry-over can be very useful.

Established methods for determining carry-over in milking systems require conducting "milkings" in a milking system with known concentrations of a tracer substance of some kind, e.g. color tracers or mixtures of artificial milk with known constituent concentrations. These methods require special equipment, possibly larger amounts of milk, and certainly also additional labor and time. Calculation of carry-over is finally done using the values for amounts of milk from subsequent milkings and values for e.g. fat content – values that are available from ICAR farm tests for certification of milk meters and samplers anyway. Therefore the aim of this study was to assess the usefulness of ICAR farm test data to estimate carry-over in milking systems.

### **Methods**

An easy way to calculate carry-over is using a linear regression model. The most basic approach uses the observation of the fat content of the current milking and tries to explain this using the fat content of the previous milking and the current milking's milk yield as regression factors:

$$f_t = \mu + a \cdot f_{t-1} + b \cdot m_t + \varepsilon \tag{1}$$

with  $f_t$  as the observed fat content f in milking t,  $\mu$  as the intercept, a as the regression coefficient for  $f_{t-1}$ ,  $f_{t-1}$  as the fat content f in milking t-1, b as the regression coefficient for  $m_t$ ,  $m_t$  as the milk yield in milking t, and å as the random residual. The regression coefficient a is the estimate of carry-over, and the regression coefficient b provides the information whether carry-over depends on milk yield or not.

However, there is more information available in an ICAR farm test that may be put to use in estimating carry-over. There can be farm effects, milk meter effects, sampler effects, or cow effects that come to mind. When tests on a farm are done with the same devices on more than one day, there also might be a day effect to consider. For use with the data available for this study, model (1) therefore was adjusted as follows:

$$f_{dt} = \mu + a \cdot f_{dt-1} + b \cdot m_{dt} + dev_{ik} + c_{jk} + \varepsilon$$
(2)

with, in addition to model (1), d denoting the milking time,  $dev_{ik}$  as the effect of device i on farm k, and  $c_{ik}$  as the random effect of cow j on farm k.

The data available for analysis included data from three AMS farm tests and one conventional milking system farm test. An overview about the available data is shown in table 1.

Using this statistical model will result in some carry-over estimates, but that does not give enough information yet. It is also important to know how reliable these results are. In this case this means that it is necessary to know if a farm test setup is able to detect a carry-over if it actually exists. In statistics this can be addressed by calculating the power of a statistical model. For more complex models like the one above it is necessary to run some simulations to estimate the test power. A test power to aim for

		Meters/	Milking		
Device	Farms	samplers	sequences	Cows	Milkings
AMS 1	2	4	8	180	256
AMS 2	1	2	2	63	74
AMS 3	1	1	2	77	120
CON 1	1	6	2	190	240

Table 1. Overview of structure of available data from ICAR farm tests.

is at least 80 % usually. A value of 50 % means that flipping a coin will lead to a result comparable with the test setup, and conducting a test in that case must be considered a waste of resources.

In the second part of this study data from an experimental farm was used to set up simulated farm tests. The original data set consisted of 16 years of data from official milk recording from a research farm, a total of 29,533 data sets from 1,108 individual cows. These data were classified per cow by lactation levels (1, 2, 3-5 and 6+) and days-in-milk levels (d" 95 d, d" 185 d, d" 305 d, and > 305 d). For each combination the mean milk yield and fat content per cow was calculated. These values were considered a pool of "cows" to select from when setting up a simulated herd, with about 7,500 individual records. This is sufficient for setting up a test with individual milking times, where every cow can only be milked once per sequence. For an AMS test setup it needs to be considered that the same cow can be milked again after a certain time. To take this into account, every milking requires having an associated milking duration, so that it can be simulated when a cow is back in the pool of cows to choose from when setting up a milking sequence for an AMS system simulation. This can be done by also giving each "cow" an average milk flow rate.

Based on this data set, the simulation process can be started. The simulation consisted of several steps, as follows:

- Choose type of device: AMS or conventional milking system. This predefines the number of devices and samplers per farm, the number of total milkings and the number of milkings per milking sequence. In an AMS system it is common to test two samplers on two farms with one AMS box each, with at least 50 valid milkings per combination. A test in a conventional milking system usually consists of testing four devices and samplers per farm, with at least 40 valid milkings per device.
- 2. Choose number of cows per farm or group. This determines how many milking sequences are necessary to get the required number of valid milkings.
- 3. Create the test herds: select the desired amount of "cows" from the aforementioned pool. Then randomly select "cows" from that herd to create a milking sequence for every milking time. In a conventional system every "cow" may only turn up once per milking time. In an AMS simulation "cows" can return to the pool after they have been selected and then waited for a minimum amount of time. In this study a minimum waiting time of 6 h was used.
- 4. Create the carry-over: Schedule a "true" carry-over for the system under test, and apply some variation for each individual milking. This can be done by using standard error estimates from the real data, for example. Then use milk yields and fat contents to calculate the "true" fat content for every milking. Based on the true fat content and the true carry-over the resulting fat content in the samples can be calculated.



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- 5. Run the statistical linear model for carry-over estimation with the simulated data set. Check if the carry-over estimate is significantly different from zero, and check the deviation of the estimated value from the true value.
- 6. Repeat steps 2 to 5 to get a good estimate for the test power, i.e. the percentage of simulation that found a significant result for the carry-over estimate. For this study 100 repetitions were made. The simulation then can be repeated for different "true" carry-over values to get a better idea about the boundary conditions for detecting carry-over with sufficient test power.

# Results and discussion

The results from the mixed linear model (2) can be found in table 2. None of the carryover estimates was significant. This may be true for AMS 1, for the other AMS systems some carry-over could be expected. Standard errors are high for the AMS systems, which indicate that it will be more difficult to detect a true carry-over. The conventional system actually has some carry-over, so the result from the linear model is likely false in this case.

Figure 1 shows the estimated carry-over values from 100 simulations for ten different "true" carry-over values ranging evenly from 2 % to 20 %, based on an ICAR test setup for an AMS. It can be seen that variation between the simulations within each carry-over value is high, likely resulting from both high initial carry-over variation as well as variation from "herd composition", selected "cows" and "milking sequences".

In figures 2 and 3 the test power estimates for various carry-over levels are shown. Low carry-over values are rarely detected properly in these test setups – the paradoxically increased number of detected carry-over-levels for the conventional milking system actually stems from negative carry-over estimates that are different from zero. All in all, a somewhat sufficient detection of existing carry-over can only be found for higher values of 16 % or higher.

In figure 4 the variation of carry-over estimates between conventional milking system setups and AMS setups can be compared. The values are slightly higher for the AMS simulation, which might be due to the lower number of devices in the AMS test setup compared to the conventional milking system. The longer milking sequences in the AMS test setup and the potential repeated measurements on the same cow being milked twice in an AMS sequence probably cannot make up for the larger number of devices in the conventional system with regard to variation between simulations.

Table 2. Results for carry-over estimates for different milking systems from ICAR farm tests. SE denotes the standard error of the carry-over estimate.

Device	Carry-over estimate	P-value (t-test)	SE
AMS 1	0.03	0.5557	0.05
AMS 2	0.15	0.0628	0.08
AMS 3	-0.01	0.8326	0.05
CON 1	0.00	0.9615	0.01





Figure 1. Results from several carry-over estimations for an AMS system ICAR test setup.



Figure 2. Test power estimates for ten different carry over levels, for a conventional milking system ICAR test setup.



Figure 3. Test power estimates for ten different carry-over levels, for an AMS system ICAR test setup.





Figure 4. Estimated carry-over value ranges, for a conventional milking system ICAR test setup (left) and an AMS system ICAR test setup (right).

It must be considered that the data from the original ICAR farm tests can be improved. For three of the four milking systems only half the amounts of data were useful, since there were no cow IDs for the other test farms available. This can lead to smaller standard errors and in consequence also to less variation in the simulations.

#### Conclusion

Based on the simulation results it is difficult to justify using data from ICAR farm tests for carry-over estimation without further adjustments. For better results more information input is necessary, especially knowing some more true carry-over levels (and their variation!) of given milking systems as reference values will be helpful. Adding data from more previous ICAR tests with potentially more than the minimum measurement requirements due to repeating tests in case the device did not pass in the first time might also be useful to get a better grasp of the ability to detect carry-over. Lastly, improving the statistical model is also necessary to avoid paradoxes like negative carry-over estimates, or to take a closer look at the dependency between carry-over and milk yield.

Overall, the critical carry-over levels to detect are in the range from 2 % to 8 % for most lab methods. At the moment this seems to be a challenge. It could be helpful to look at other traits like protein and lactose content in addition to fat content to improve estimation, or put some thought into alternative ways of estimating or measuring carry-over.