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SmartCow: an integrated infrastructure for increased research capability and innovation in the European cattle sector

SmartCow
an integrated infrastructure for increased research capability and innovation in the European cattle sector

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EXECUTIVE SUMMARY

Background	In the frame of SmartCow project, SRUC has provided access to its Research Installation "Dairy Research and Innovation Centre" through Trans National Access (TNA).
Objectives	This Deliverable aims at describing the TNA provided by SRUC Dairy centre during the SmartCow project.
Methods	The Deliverable is composed of a table summarising the TNA provided by the Research Installation (RI) and by the reports of activities provided by the TNA users who accessed this RI.

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1 TNA provided

Name of the TNA project	Name of TNA user	Organisation of TNA user	Country of TNA user	Installation from the RI	Start date	End date	Number of units of access provided
VitalCow – Skin carotenoid sensor	Martina Jakob	Leibniz Institute for Agricultural Engineering and Bioeconomy	Germany	SRUC Dairy Centre	17/2/20	30/08/20	100 cow-weeks
Novel trace mineral source	Denise Cardoso / Stephane Durosoy	Animine SA	France	SRUC Dairy Centre	22/10/21	16/12/21	192 cow-weeks
Amino acid nutrition	Lahlou Bahloul	Adisseo SA	France	SRUC Dairy Centre	21/2/22	30/4/22	210 cow-weeks

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2 Final reports of the each TNA provided

2.1 TNA 1: Martina Jakob (Leibniz)

Regular health monitoring is necessary to improve the longevity of modern, high producing dairy cows. Health problems, such as mastitis, are one of the main reasons for culling on dairy farms globally. Early detection of diseases can help minimize the need to cull and improve overall animal health, welfare, and production Mastitis is commonly detected by identifying increased somatic cell count in milk, via California Milk Test, milk electrical conductivity changes or lactate dehydrogenase activity. However, most of these approaches are not suitable, or reliable enough, for early detection. A disease specific approach to detection could overcome this. For example, identification of inflammatory processes associated with mastitis at an earlier stage could allow for earlier detection, reducing animal welfare impacts, production impacts and financial impacts and reducing the need for antibiotic use on farm. The aim of this pilot study was to determine whether it is possible to accurately measure skin carotenoid content of dairy cows, as a proxy for health status, by measuring teat and udder skin with a commercially available multiple spatially resolved reflection spectroscopy (MSRRS) sensor intended for human use.

Hypothesis tested:
It is possible to use a sensor intended for use in humans to measure skin carotenoid levels of lactating dairy cattle via teat skin measurements as a proxy for inflammatory diseases, such as mastitis.

Material and Methods:
Thirty-two randomly chosen lactating Holstein-Friesian cows with variations in yield, stage of lactation and parity were measured as part of this trial with the only selection factor being little pigmentation on the teats. Measurements were collected once daily, prior to afternoon milking. Repeat measurements were taken from two teats, ensuring the sensor was not moved between repeats. A total of 2080 measurements were collected. Animals were housed in loose cubicle housing with no outdoor access and maintained on a total mixed ration (TMR) formulated for maintenance + 30L, consisting of homegrown forages (grass silage, whole crop and maize silage) and concentrate ingredients, balanced nutritionally with minerals. Measurements started in March 2020 with an initial group of 20 animals and stopped after ten days as a result of an enforced lockdown due to the covid-19 pandemic. The trial continued in July 2020 with another 13 days of measuring. The same cows were utilized as far as was possible, considering stage of lactation and drying periods. Where necessary, animals were replaced with those as similar as possible.

Measurements were taken manually per cow by placing the sensor firmly on the teat skin, ensuring the full sensor surface was covered. Once the sensor was fully covered, the measurement started automatically and light emission for a complete cycle took approximately 15 seconds. During this time, the sensor was kept still until the software signaled the end of the measurement. Sensor position (teat, or side of the udder), cow identification number and time of measurement were manually recorded. Sensor data was then extrapolated to give a carotenoid content value in the skin sampled, based on a self-developed algorithm by Opsolution (Germany). Additional animal data were collected from sites, including dietary information, breed, body weight, body condition score (BCS), stage of lactation (days in milk), milk yield and lactation number. Using days in milk (DIM), animals were grouped via their nutritional energy balance (NEB), where between 1 and 84 DIM was considered negative EB, between 85 and 245 DIM was considered neutral and above 245 DIM was considered a positive EB. BCS was carried out by one observer utilizing a 21-point scale based on NIRD dairy cow condition scoring, with quarter point scores. Animals were scored using direct

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physical assessment standing in a cubicle or crush. Across all sites there was one case of clinical mastitis and no other diseases were documented.

The main scientific outcome, innovation/impact of the results:
Overall, the measurement quality in Dumfries was excellent, with nearly 75% achieving the highest quality possible. A decrease in quality was mostly coupled with a decrease in the value displaying the carotenoid content. The average value for all cows in Scotland was 10.7. The French average value, based on 2467 samples, was 14.1 and the Irish average, based on 468 samples was 9.4. The carotenoid content of humans and cows appears to be similar, but cows have a slightly higher value, as expected given their diet being high in forages. The values of the French sample exceeded the sensor's calibrated range of 15, highlighting the limitation of using a sensor calibrated for human skin.
Overall, the dynamics of carotenoids are little researched for dairy cows. For the French sample, a statistically significantly higher sensor value was associated with those animals which were grazed. Fresh grass and sunlight can increase the level of carotenoids in the skin. Nutrition is generally the main influencing factor on the carotenoid content in the skin, and it is therefore plausible to observe a raise in the value between fresh grass and silage. There were also significant differences between breeds which may be explained by variations in milk yield, which could affect oxidative stress– though there was no significant relationships observed with nutritional energy balance.
There was only one incidence of mastitis in all samples. The affected cow showed a slight drop in the value (-1), but the day the mastitis was diagnosed was not measured as it was a weekend, and on weekends, no measurements took place. Therefore, the acute phase of the mastitis was missed. Two days after the start of the antibiotic treatment the value was at the same level as it was before the mastitis.
The achieved results show that the sensor has potential to be used in cows to measure carotenoid content in skin. Further trials with contemporaneous blood analysis for the blood carotenoid content of the cows are needed now, to adjust calibration for bovine skin.

How do you expect to disseminate the results:
Part of the results were presented at a conference in September 2020, but the Scottish sample was not included:
(https://www.agroscope.admin.ch/agroscope/de/home/aktuell/veranstaltungen/akal2020.html)

The full 3 TNA studies (Scotland, Ireland, France) have been combined into a manuscript and are currently (April 2022) under peer review in Journal of Dairy Science Communications as "Skin carotenoid levels in lactating dairy cows as measured using multiple spatially resolved reflection spectroscopy".

2.2 TNA 2: Denise Cardoso / Stephane Durosoy (Animine SA, France)

Experiments conducted *in vitro* have provided evidence for ruminal responses (e.g., acetate:propionate ratio) to Animine's proprietary potentiated zinc oxide (Fellner et al., 2021, Applied Animal Science 37: 27–32), compared with other sources of Zn. Whether these responses are relevant *in vivo* is not known. The objective of this study was to determine the effect of a mix of Animine proprietary sources of Zn and Mn on cow performance (milk yield and composition, body

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weight and condition score) and – potentially - rumen function (pH and VFA concentrations) and total tract digestibility in lactating dairy cows.

Hypothesis

Providing Zn and Mn as Animine proprietary sources rather than sulphates will increase ruminal acetate:propionate ratio, increase total tract NDF digestibility and increase yields of milk protein and fat.

Material and Methods

The experimental design was based on that used by Faulkner and Weiss (2017, J. Dairy Sci. 100: 5358–5367) to compare hydroxy trace minerals with sulphates in lactating dairy cows. Those authors used 18 cows in a split-plot Latin Square to compare two sources of Zn, Mn and Cu at two levels of dietary fibre, using 28d periods. In our experiment, two sources of Zn and Mn were evaluated using a 2 x 2 crossover design, with 28d periods and 24 lactating Holstein cows less than 150 days in milk arranged in 12 blocks (pairs) according to parity (primiparous or multiparous) and stage of lactation. We did not include Cu due to the known high Cu status of the SRUC herd (based on forage Cu concentrations and liver Cu measured in cull cows).

Within pairs, one cow was allocated at random to either treatment sequence AB or treatment sequence BA and the second cow was allocated to the treatment sequence not followed by the first cow.

Treatments were:

A. Control. Zn and Mn provided as sulphates.
B. Animine. Zn and Mn provided as proprietary products 'HiZox', 'CoRouge' and 'ManGrin' (Animine SA, France)

In both cases, diets were formulated to supply 75% of the Zn and Mn permitted by EU (and UK) feed legislation.

Cows were housed in a single group in a cubicle (free-stall) barn and offered the same total mixed ration (TMR) for *ad libitum* intake via electronically operated Hokofarm feeding stations (Insentec, Netherlands). Cows had free access to fresh drinking water at all times. Trace minerals were provided within manufactured premixes designed for use within TMR.

The TMR was formulated using the 'Feed into Milk' system to 12.4 MJ ME/kg DM and (as % DM) 16.6, 33.1 and 14.9 CP, NDF and starch, respectively. Forages were grass silage and whole crop wheat silage at 38.0 and 17.6% of DM, respectively.

A manufactured vitamin/mineral premix was included at 0.7% of diet DM (targeting an intake of 150g/cow/d). Premixes contained 10250mg/kg Zn as zinc sulphate monohydrate (treatment A) or as zinc oxide (treatment B), and 8800mg/kg Mn as manganese sulphate monohydrate (treatment A) or as manganese (II) oxide (treatment B).

Dry matter intake was calculated daily for each cow from individual meals recorded by the Hokofarm feeding equipment. Milk yield was recorded at each milking (cows were milked twice daily at 04:00 and 16:00) and concentrations of fat, crude protein, lactose, urea, somatic cells and

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major fatty acids in milk were determined by mid-infrared spectroscopy on samples collected during the last three days of each period.

Cows were weighed when entering the milking parlour and Body condition score (BCS) was estimated by two operatives at the end of each period.

Rumen contents were sampled by naso-oesophageal intubation on one occasion per period for analysis of pH, VFA and ammonia-N, with sub-samples retained (at -80 C) for possible microbiome analysis.

Faecal samples were collected at intervals on the last two days of each period to permit later calculation of total tract digestibility (using acid insoluble ash as an internal marker), depending on cow production results.

Data were analysed by analysis of variance (Genstat 18, VSN International Ltd.), fitting terms for block, period, treatment and their interactions.

The main scientific outcome, innovation/impact of the results:

On average, premixes contained more Mn (+13%) and less Zn (-5.6%) than targeted. Premix B (test) contained 5% more Mn and 9% more Zn than premix A (control).

Overall levels of cow performance met expectations: dry matter intake close to 24kg/cow/d and mean milk yield of 40.1kg/d.

Treatment had no effect (P>0.1) on any measure of cow performance (dry matter intake, milk yield, milk composition, body weight or body weight change). Due to a technical problem with the gas chromatograph, ruminal VFA concentration data are considered unreliable. Due to a change in laboratory staff, a repeat analysis of stored back-up samples has not yet been completed. No decision has yet been made concerning analysis (not funded through Smart Cow) of faecal samples for calculation of total tract digestibility.

Lack of response to treatment might be due to (a) imprecise delivery of targeted concentrations of Zn and Mn, (b) insufficient experimental power (replication was based on the precedent offered by Faulkner and Weiss, 2017), or (c) targeted concentrations of Zn and Mn were insufficient to cause, *in vivo*, effects previously observed *in vitro*.

How do you expect to disseminate the results?

Results may be reported at conferences such as RRR (France) or ADSA (USA).

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2.3 TNA 3: Lahlou Bahloul (Adisseo SA, France)

Amino acid nutrition affects both dairy cow productivity (e.g., milk protein yield) and the environmental sustainability (e.g., nitrogen use efficiency, NUE) of milk production. More precise diet formulation to optimise the profile of essential amino acids in metabolisable protein (MP) may permit use of lower protein diets without loss of milk protein production, thereby increasing NUE.

While considerable research is available documenting responses to improved amino acid nutrition (through the use of rumen-protected amino acids), responses of high-yielding cows offered low protein diets based on extensively fermented grass silage (a common production system in NW Europe) are poorly characterised. Our objective was to fill this knowledge gap.

Hypotheses

1. Compared with the current commercial diet (based on extensively fermented grass silage), reducing dietary CP and MP without optimising amino acid profile will reduce productivity (milk protein yield) and have no effect on environmental sustainability (NUE).

2. Optimising the amino acid profile of a diet containing lower than current commercial CP and MP will improve productivity (milk protein yield) and improve environmental sustainability (NUE).

Material and Methods

Three treatment diets were compared using a randomised complete block design, using 63 multiparous lactating Holstein dairy cows less than 150 days in milk. Cows were first arranged into three blocks (cohorts) of 21 based on calving date. Each cohort occupied the experimental facility consecutively for 35d. *Cohorts 1 and 2 completed the study before 30 April 2022 (i.e., within the Smart Cow project). Cohort 3 cows are scheduled to be on experiment in May and June 2022, supported by additional funding directly from Adisseo SA.*

Treatments were:

A. Total mixed ration (TMR) formulated to meet ME and MP requirements according to the Feed into Milk model (Thomas, 2004) (as a diet descriptor, CP = *circa* 17.5% CP in DM).

B. Lower protein TMR, formulated to meet ME requirements and 0.95 of MP requirements (diet is expected to be *ca.* 16% CP in DM).

C. Amino acid optimised TMR, formulated to meet ME requirements and requirements for metabolisable methionine and lysine. This diet will be isonitrogenous with diet B.

63 multiparous lactating Holstein dairy cows less than 150 days in milk were first arranged into three blocks (cohorts) of 21 based on calving date. Each cohort occupied the experimental facility consecutively. Within each cohort, cows were arranged into seven blocks of three according to stage of lactation and milk protein yield. Within each block of three, cows were allocated at random to treatment and offered treatment diets for 35d.

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Diet ingredients were grass silage, whole crop wheat silage, dried sugar beet pulp, rapeseed meal, rumen-protected rapeseed meal, maize distillers' grains, rumen-inert fat, vitamin/mineral premix and a source of rumen-protected methionine.

Cows were milked twice daily, feed intake was recorded daily (from visits of individual cows to Hokofarm feed bins) and milk composition (fat, crude protein, lactose, urea, somatic cells and major fatty acids) was measured by mid-infrared spectroscopy in samples collected throughout the 35d experimental period. Cows were weighed automatically on entry to the milking parlour and Body Condition Score was recorded at the beginning and end of the experimental period.

The main scientific outcome, innovation/impact of the results:

Results are presented for cohorts 1 and 2, pending complete data and analysis for cohort 3.

Feed analysis

For cohort 1, concentrations of CP in TMR (Table 3) were 4, 4, and 3% less than targeted for TMR A, B and C, respectively (Table 1). Measured CP in TMR was similarly below target for cohort 2.

Table 1. Analysis of total mixed rations

	Cohort 1			Cohort 2		
	TMR A	TMR B	TMR C	TMR A	TMR B	TMR C
DM (g/kg)	450	454	447	435	429	428
CP (g/kg DM)	174	155	157	174	151	155
NDF (g/kg DM)	314	345	341	355	375	371
Starch (g/kg DM)	182	168	160	156	170	167
Ash (g/kg DM	77	80	80	74	71	73
Oil (g/kg DM)	50	52	53	52	52	51

These shortfalls in CP can be attributed to lower than expected CP concentrations in blends and grass silage (data not shown).

Cow performance

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Mean results across cohorts 1 and 2 are presented: differences between cohorts will be evaluated when full results are available for cohort 3.

Treatment groups did not differ in the stage of lactation of cows at enrolment to the study (103, 107 and 113 days after calving for treatments A, B and C, respectively (P>0.05)).

Treatment had no effect on DMI (Table 4). However, cows on treatment A were lighter than those on treatments B and C, such that DMI as % BW was lowest for treatment B and highest for treatment A.

Milk yield tended to be lower for the two low protein treatments, B and C, than treatment A, although the difference was numerically large (4.7kg/d). Consistent with this, milk urea concentration was lower for treatments B and C than treatment A.

Milk macro-composition (lactose, fat and crude protein concentrations) was not affected by treatment, while milk urea was significantly lower for the two low protein treatments, B and C, than treatment A.

Largely as a consequence of differences in milk yield, yields of lactose, fat and protein all fell when the concentration of dietary protein was reduced. This fall in N output compensated for the reduction in N intake, such that NUE was not affected by the reduction in dietary CP.

No cow performance variable was different between treatments B and C.

Table 2. Cow performance.

	A	B	C	Sem	P
DMI (kg/d)	23.7	23.5	23.7	0.59	0.945
DMI (% BW)	3.46[a]	3.17[b]	3.35[ab]	0.082	0.057
Milk yield (kg/d)	39.5	34.8	34.7	1.53	0.053
Milk lactose (%)	4.61	4.51	4.49	0.041	0.103
Milk fat (%)	4.27	4.18	4.23	0.149	0.908
Milk crude protein (%)	3.19	3.16	3.19	0.079	0.957
Milk urea (%)	0.0155[a]	0.0062[b]	0.0073[b]	0.00199	<0.001
Somatic cell count (Kcell/ml)	30	105	104	39.1	0.310
Milk lactose (g/d)	1822[a]	1566[b]	1556[b]	71.4	0.020
Milk fat (g/d)	1680[a]	1450[b]	1436[b]	62.8	0.016
Milk protein (g/d)	1255[a]	1096[b]	1077[b]	40.9	0.008
NUE	0.299	0.298	0.286	0.086	0.477
Body weight (kg)	686[a]	743[b]	711[ab]	12.7	0.012
Body weight change (kg/d)	0.064	0.150	0.047	0.1163	0.799
Body Condition Score	*Data not yet evaluated*				

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BCS change (units/d)	*Data not yet evaluated*

Discussion

Lowering the concentration of dietary CP (treatment B versus treatment A) reduced milk protein yield but did not increase NUE.

Responses of milk protein yield to MP are curvilinear, with the apparent efficiency of utilisation of MP for milk production approaching 0.64-0.67 at low protein intakes (well below estimated requirements) and declining to typically 0.15 or lower as protein intake increases.

In this experiment, across all diets, measured dietary CP was ca. 7-8g/kg DM lower than target levels, placing the diets on a steeper part of the response curve than planned. Thus, the decline in milk protein yield is expected to be higher, and the gain in NUE lower, than anticipated.

Using formulated values for MP concentration (Table 2) and treatment means for milk protein yield (Table 3), and assuming no difference in the partitioning of MP, the calculated marginal efficiency of utilisation of MP for milk protein production is actually greater than 1. Body weight change was not different between treatments, so a major contribution to MP supply from mobilised body protein is unlikely. Perhaps the most likely explanation is that the difference between treatments in MP was greater than the 4g/kg DM (108-104) estimated when diets were formulated. The contribution of MP from digestible undegradable protein (DUP) may have been less than planned. Further calculations of the N economy for individual cows is warranted.

Adding methionine in the form of Metasmart Dry did not increase milk protein yield or NUE (treatment C versus treatment B). It is possible that other nutrients (such as metabolisable lysine or histidine) were limiting to milk protein synthesis. The possibility of degradation of the active molecule in Metasmart (HMBi) under the damp conditions of the TMR is currently being investigated.

How do you expect to disseminate the results?

Results may be reported at animal science conferences such as RRR (France), BSAS (UK) or ADSA (USA).

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