

Veterinary diagnostics on farm - a review

J. Hakker¹ and C.A. Makkink¹

¹Kepto, P. O. Box 2081, Deventer, The Netherlands

Email: j.hakker@kepto.nl; c.makkink@hetnet.nl

SUMMARY

Good management, antibiotic resistance awareness, and biosecurity are increasingly important in dairy farming. One of the biggest challenges for a livestock veterinarian, however, is to educate and convince farmers to change management instead of using drugs to mask mistakes in their management. The confidence of the farmers and the willingness to adjust their management practices may increase if the veterinarian is able to demonstrate the possibilities for improvement on farm. Improvement of farm management can be achieved by the use of on-farm diagnostic tests. On-farm diagnostic tests make early detection of diseases possible and reduce inappropriate use of antibiotics. Examples of on-farm tests are the ATP meter for monitoring of drinking water quality, the California mastitis test for detection of subclinical mastitis and the ruminal pH test for investigation of feed management. The use of these tests may change the attitude of the dairy cattle farmer from curative thinking towards more preventive thinking.

Key words: antibiotic resistance, on-farm tests, diagnostics, management

INTRODUCTION

Veterinary diagnostics on farm is an important aspect of veterinary science. Analysis of symptoms and determining the correct diagnosis are the first steps in the development of a successful treatment plan, the prevention of disease spreading, and improvement of general health status. In 2017, Overton *et al.* published an extensive review on the development of metabolic health indicators over the past century (Overton *et al.*, 2017). They concluded that the development of on-farm and cow-side diagnostics has come a long way, and will proceed into real-time and automatic monitoring of cattle health on dairy farms.

Monitoring health status on farms is becoming more and more important, because early diagnosis of health problems enables a farmer to act in the early stages of a health issue, which will help to reduce the use of drugs and the development of resistance against antibiotics. Antimicrobial resistance, induced by excessive use of antibiotics, is considered a serious threat to public health (Ferri *et al.*, 2017). For effective health monitoring it is important that the farmer is motivated, and that it takes not much time and

effort to collect relevant data (Egger-Danner *et al.*, 2015).

On-farm diagnostics

Enabling dairy cattle farmers to keep a close eye on the health status of their herd is crucial for preventive and curative on-farm health care. Products for diagnosing health issues and disease in livestock are widely available worldwide. These products for on-farm, cow-side diagnosis are usually offered in the form of test kits. It is of crucial importance that such kits offer reliable outcomes to enable the farmer to adjust health management and disease control measures on his farm according to the test results. Tatone *et al.* (2016) point to the fact that meta-analysis of diagnostic test accuracy is more and more common in human medical literature, but few veterinary examples of such meta-analyses are available (Tatone *et al.*, 2016).

On a dairy farm, next to urine, manure and blood, milk is an easily available substance to sample and analyse for the purpose of health monitoring. Milk composition is already used to monitor metabolic status and energy balance of dairy cows (Egger-Danner *et al.*, 2015). The number of commercially offered tests for analysis of milk samples for disease or pregnancy diagnosis is

expected to increase (Barkema *et al.*, 2015). Electronic devices, measuring physical activity or body temperature, may be applied on farm to detect oestrus or fever in dairy cows (Egger-Danner *et al.*, 2015). Neethirajan (2016) also sees emerging markets for wearable biosensors to be applied on dairy farms for the continuous monitoring of animal health. From his extensive overview it is concluded that the role of wearable biosensors in dairy farming will increase in the future, and that these technologies will help to create real-time monitoring of herd health status, including early diagnosis of diseases.

Diagnostic tools for the detection of sub-clinical ketosis in dairy cattle

Tatone *et al.* (2016) performed a comparative study on the accuracy of tests for detecting hyperketonaemia in dairy cattle. They concluded that diagnosing hyperketonaemia based on milk, urine or blood testing are all valid options, however, urine and milk samples yield more diagnostic inaccuracy than blood samples. Handheld meters to measure beta-hydroxybutyrate levels in blood serum or plasma are reliable devices to detect sub-clinical ketosis in dairy cows (Voyvoda and Erdogan, 2010; Pineda and Cardoso, 2015).

Diagnostic tools for the detection of sub-acute rumen acidosis in dairy cattle

Sub-acute rumen acidosis (SARA) is a frequently occurring health issue in dairy cattle in early and mid-lactation: up to one quarter of the animals in a dairy herd may suffer from SARA, which is caused by accumulation of volatile fatty acids, and insufficient buffering capacity (Kleen *et al.*, 2003; Plaizier *et al.*, 2008). Improper transition management and/or a too sudden change of ration composition around calving may trigger the disease (Kleen *et al.*, 2003). Problems related to ruminal pH depression may be related to the ration, i.e., feeding excessive amounts of non-structural carbohydrates and highly fermentable forages, and insufficient dietary coarse fiber (Kleen *et al.*, 2003; Plaizier *et al.*, 2008). SARA may result in a decrease in feed intake, rumination, fiber fermentation, and milk fat content. More serious symptoms include diarrhea,

laminitis, parakeratosis of the rumen wall, liver abscesses; increased production of bacterial endotoxin and inflammation characterized by increases in acute phase proteins (Kleen *et al.*, 2003; Plaizier *et al.*, 2008). It is clear that early diagnosis of SARA is of utmost importance to maintain a high health status on a dairy farm. Usually, depressed milk fat content is a practical tool for the detection of SARA in mid-lactation (Enemark, 2008). Another option is measuring the pH of rumen fluid. Garrett *et al.* (1999) found that the ration composition of dairy cattle (low or high forage content) was reflected in the rumen pH. Not many techniques are available for measuring rumen pH under field conditions. Duffield *et al.* (2004) name two options: rumenocentesis and oral stomach tube. They concluded that rumenocentesis (sampling fluids directly from the rumen) was more sensitive and more accurate than pH measurements based on the oro-ruminal probe. When taking rumen fluid samples by oro-ruminal tube, contamination with (alkaline) saliva is difficult to avoid, resulting in inaccurate pH assessment. In his review article on SARA, Enemark (2008) also advocates rumenocentesis as the most reliable tool for diagnosing SARA.

The authors also emphasize that improved field techniques are required for better on-farm diagnosis of SARA, possibly with the aid of sensors that continuously monitor rumen pH (Duffield *et al.*, 2004; Enemark, 2008). Such a sensor (Farm Bolus) was used by Villot *et al.* (2017) to monitor rumen pH continuously. The authors conclude that relative rumen pH indicators are useful tools to be implemented in precision livestock farming devices (rumen boluses) to detect digestive disorders in cows at herd level (Villot *et al.*, 2017).

Diagnostic tools for the detection of (sub)clinical mastitis in dairy cattle

Somatic cell count (SCC) in milk is widely used to assess udder inflammation in dairy cows. The California mastitis test provides a simple cow-side indicator of subclinical mastitis, by detecting bacterial DNA in milk samples. To select adequate control measures, however, it is crucial to identify the causative pathogen. In 2015, Duarte *et al.* published an extensive overview of

recent advances in bovine mastitis diagnosis. They state that the demand for fast and reliable diagnostic procedures will continue to rise as farm size in the dairy industry continues to increase. Duarte *et al.* (2015) expect that new technologies - such as transcriptome and proteome analyses and nano- and microtechnology in portable devices - will offer sensitive, practical and reliable methods for the detection of mastitis pathogens and inflammation biomarkers. Ashraf and Imran (2018) also recognize the need for new methods for the early diagnosis of mastitis on dairy farms. They see traditional methods, like SCC and microbial culturing, being replaced by modern technologies, like polymerase chain reaction and sequencing-based tests. Future cow-side tests, incorporated in biosensors, may be based on nanotechnology and protein-based technology, according to Ashraf and Imran (2018).

Griffioen *et al.* (2016) interviewed almost 200 Dutch dairy farmers about their attitude towards on-farm diagnostic tests for mastitis. For dairy farmers, obtaining reliable and fast results were the most important criteria when choosing a microbiological mastitis test method. Increasing the availability of such tests will, therefore, stimulate the use of on-farm test methods, and, consequently, optimize antibiotics use in dairy farmers, the authors state.

Diagnostic tools for the detection of diarrhoea in calves

Neonatal diarrhoea is the main cause of mortality and growth depression in young calves. Diarrhoea may be caused by bacteria, viruses, or parasites, or by combinations of pathogens. The multifactorial nature of calf diarrhoea makes this disease hard to control effectively in modern cow-calf operations (Cho and Yoon, 2014; Muktar *et al.*, 2015). Common pathogens involved in neonatal calf diarrhoea are rota viruses, corona viruses, *E. coli*, *Salmonella*, *Campylobacter*, *Clostridium* and *Cryptosporidia* (Fournier and Naciri, 2007; Cho and Yoon, 2014; Muktar *et al.*, 2015). It may be difficult to distinguish between the various possible causes, but a quick and reliable diagnosis is crucial for a correct treatment plan (Cho and Yoon, 2014). Various laboratory methods are used to identify enteric pathogens,

but not many methods are applicable in the field (Cho and Yoon, 2014; Muktar *et al.*, 2015).

For the identification of bovine coronavirus and type A rotavirus, diagnostic methods based on direct electron microscopy (DEM), enzyme-linked immunosorbent assay (ELISA), and protein A-gold immunoelectron microscopy (PAG-IEM) are available (Athanasious *et al.*, 1994). All three methods can be effectively employed in a centralized diagnostic laboratory, but the authors recommend to use PAG-IEM to verify doubtful findings. The three methods evaluated by Athanasious *et al.* (1994) are not suitable for routine on-farm application. Papini *et al.* (2018) evaluated three commercially available immunochromatographic test kits to detect *Cryptosporidium parvum* in calf diarrhoeic calf stool. They concluded that all three tests (FASTestV R CRYPTO strip, FAS TestV R CRYPTO-GIARDIA Strip and TETRASTRIPSV R) were easy to perform and yielded reliable results. A practical field test to diagnose the cause of neonatal diarrhoea in young calves is the Speed V-Diar TM test, which is also applied on calf faeces. Neonatal calf scouring is often related to insufficient colostrum intake or inadequate colostrum quality (Cho and Yoon, 2014; Muktar *et al.*, 2015).

Tools for assessing colostrum quality

Fortunately, colostrum quality can easily be monitored on-farm. Well-known tools for this are the colostrometer and the Brix refractometer. Bartier *et al.* (2015) compared both methods and concluded that although the colostrometer data are better correlated with true IgG values (as determined by radial immunodiffusion), the user-friendly Brix refractometer is a more specific tool to detect colostrum of adequate quality. Biemann *et al.* (2010) evaluated optical and digital Brix refractometry instruments, and they concluded that both instruments yielded excellent results.

Tools for the monitoring of farm health and hygiene

Apart from specific tests for the early diagnosis of various livestock diseases, general health and hygiene status should also be monitored

continuously on dairy farms. This monitoring may include testing of water, feed and air quality in the barn. In water systems, biofilms consisting of bacteria and moulds may be formed, and these may pose a health risk to livestock. The microbial quality of drinking water can easily be tested by means of an ATP meter (Vang, 2013). This ATP assay measures and quantifies the active biomass in water systems, and offers quick and reliable results (Vang, 2013). The method is suitable for continuous online monitoring of water quality. On-line bacteria sensors are a novel technology, capable of distinguishing and quantifying bacteria and particles in drinking water (Højris *et al.*, 2016). Simões and Dong (2018) evaluated the use of a fluorescent optofluidic sensor for the detection of pathogens in drinking water. The method is based on tryptophan intrinsic fluorescence, and offers a low-cost solution for monitoring water quality.

Air pollution is a well-known cause of human health issues. Also in dairy farming, the concentrations of ozone, nitrogen dioxide, and particulate matter may be detrimental to cow health (Cox *et al.*, 2016). It may, therefore, be useful to develop simple, automated methods for monitoring air quality in cattle barns. Cox *et al.* (2016) emphasize that weather conditions, especially high temperatures and relative humidity, may interfere with air quality. The quality of the ration of dairy cattle is determined largely by the quality of forages, since the composition and quality of concentrates is monitored and guaranteed by the supplier. Silages and other forages can be easily inspected by the farmer, based on sight and smell. Also, visual inspection of manure will provide insight in digestive efficiency. Ration components and totally mixed rations may be tested for their composition in a nutrition laboratory.

Future of on-farm diagnostics in livestock

Apart from state-of-the-art diagnostic technology for use in veterinary laboratories, it is also very important to further increase the availability of fast, easy applicable, cheap and reliable on-farm-diagnostics. Even if adequate on-farm test kits are available, it may be necessary to persuade farmers to make use of these possibilities: Vande Velde *et*

al. (2015) recommend to use theories from the field of psychology, e.g., the Theory of Planned Behaviour and the Health Belief Model, to develop communication strategies to advocate sustainable disease control - including cow-side diagnostic tools - on dairy farms.

On farm screening of health status and confirmatory tests are important parts of general disease control in livestock farming. The development of easy-to-use test kits based on immunological, molecular, chemical and genetic diagnostics is likely to proceed in the future (Dahlhausen, 2010; Bonkobara, 2016; Ashraf and Imran, 2018). Also, the use of biosensors will enable data to be gathered and stored automatically, without additional labour required from the farmer. This provides the dairy farmer with an abundance of information, and facilitates the on-farm herd health management.

Conclusion

Maintaining good dairy herd health requires proper management and especially continuous monitoring of animal health and production parameters. Prevention, early diagnosis, and adequate treatment are key factors in disease control. In an early stage of any health issue, management or nutritional measures are often sufficient to counteract the problems. Treatment with antibiotics or other drugs is in many cases not needed, as long as the farmer and his veterinarian work together to optimize herd health status. On-farm diagnostic tests enable the farmer and the vet to keep a close eye on animal health on the farm. In conclusion, fast and reliable on-farm diagnostics contribute to the reduction of drug use in dairy farming.

REFERENCES

- Ashraf A, Imran M.. Diagnosis of bovine mastitis: from laboratory to farm. *Trop Animal Health Prod*, 50(6):1193-1202, 2018.
- Athanassious R, Marsolais G, Assaf R, Dea S, Descôteaux J-P, Dulude S, Montpetit C. Detection of bovine coronavirus and type A rotavirus in neonatal calf diarrhea and winter dysentery of cattle in Quebec: Evaluation of three diagnostic methods. *Canadian Vet J*, 35:163-169, 1994.

- Barkema HW, von Keyserlingk MAG, Kastelic JP, Lam TJM, Luby C, Roy J-P, LeBlanc SJ, Keefe GP, Kelton DF. Invited review: Changes in the dairy industry affecting dairy cattle health and welfare. *J Dairy Sci*, 98(11):7426-7445, 2016.
- Bartier AL, Windeyer MC, Doepel L. Evaluation of on-farm tools for colostrum quality measurement. *Journal of Dairy Science* 98 (3):1878-1884, 2015.
- Bielmann V, Gillan J, Perkins NR, Skidmore AL, Godden S, Leslie KE. An evaluation of Brix refractometry instruments for measurement of colostrum quality in dairy cattle. *J Dairy Sci*, 93(8):3713-3721, 2010.
- Bonkobara M. Recent developments in veterinary diagnostics: Current status and future potential. *The Vet J*, 215: 1-2, 2016.
- Cho Y-I, Yoon K-J. An overview of calf diarrhea - infectious etiology, diagnosis, and intervention. *J Vet Sci*, 15(1): 1-17, 2014..
- Cox B, Gasparrini A, Catry B, Fierens F, Vangronsveld J, Nawrot TS. Ambient air pollution-related mortality in dairy cattle: does it corroborate human findings? *Epidemiology*, 27(6):779-786, 2016.
- Dahlhausen B. Future veterinary diagnostics. *J Exotic Pet Med*, 19(2):117-132, 2010.
- Duarte CM, Freitas PP, Bexiga R. Technological advances in bovine mastitis diagnosis, an overview, 2015. *J Vet Diagn Invest*, 27(6):665-672, 2015.
- Duffield T, Plaizier JC, Fairfield A, Bagg R, Vessie G, Dick P, Wilson J, Aramini J, McBride B. Comparison of techniques for measurement of rumen pH in lactating dairy cows. *J Dairy Sci*, 87(1):59-66, 2004.
- Egger-Danner C, Cole JB, Pryce JE, Gengler N. Invited review: Overview of new traits and phenotyping strategies in dairy cattle with a focus on functional traits. *Animal*, 9(2):191-207, 2015..
- Enemark JMD. The monitoring, prevention and treatment of sub-acute ruminal acidosis (SARA): A review. *The Vet J*, 176(1):32-43, 2008.
- Ferri M, Ranucci E, Romagnoli P, Giaccone V. Antimicrobial resistance: A global emerging threat to public health systems. *Critical Rev Food Sci Nutr*, 57(13):2857-2876, 2017.
- Fournier R, Naciri M. Prévalence des agents de diarrhées chez le jeune veau. *Point Vété*, 273: 58-63, 2007..
- Garrett EF, Pereira MN, Nordlund KV, Armentano LE, Goodger WJ, Oetzel GR. Diagnostic methods for the detection of subacute ruminal acidosis in dairy cows. *J Dairy Sci*, 82(6):1170-1178, 1999.
- Griffioen K, Hop GE, Holstege MMC, Velthuis AGJ, Lam TJGM. Dutch dairy farmers' need for microbiological mastitis diagnostics. *J Dairy Sci*, 99(7):5551-5561, 2016.
- Højris B, Boesgaard SC, Albrechtsen H-J, Smith C, Dahlqvist M, A novel, optical, on-line bacteria sensor for monitoring drinking water quality. *Scientific Reports* 6:23935. Doi: 10.1038/srep23935, 2016.
- Kalorama Information,. The World Market for Veterinary Diagnostics. Market research report, 191 pages, Pub ID KLI15785446, 2018.
- Kleen JL, Hooijer GA, Rehage J, Noordhuizen JP. Subacute ruminal acidosis (SARA): a review. *J Vet Med A Physiol Pathol Clin Med*, 50(8):406-414, 2003.
- Muktar Y, Mamo G, Tesfaye B, Belina D. A review on major bacterial causes of calf diarrhea and its diagnostic method. *J Vet Med Anim Health*, 7(5):173-185, 2015.
- Neethirajan S. Recent advances in wearable sensors for animal health management. *Sensing and Bio-Sensing Research* 12:15-29, 2016.
- Overton TR, McArt JAA, Nydam DV. A 100-year review: Metabolic health indicators and management of dairy cattle. *J Dairy Sci*, 100(12): 10398-10417, 2017.
- Papini, R., Bonelli F., Montagnani M. & Sgorbini, M.. Evaluation of three commercial rapid kits to detect *Cryptosporidium parvum* in diarrhoeic calf stool. *Italian J Anim Sci*. <https://doi.org/10.1080/1828051X.2018.1452055>, 2018.
- Pineda A, Cardoso FC. Technical note: Validation of a handheld meter for measuring β -hydroxybutyrate concentrations in plasma and serum from dairy cows. *J Dairy Sci*, 98(12): 8818-8824, 2015..
- Plaizier JC, Krause DO, Gozho GN, McBride BW. Subacute ruminal acidosis in dairy cows: The physiological causes, incidence and consequences. *The Vet J*, 176(1):21-31, 2008.
- Tatone EH, Gordon JL, Hubbs J, LeBlanc SJ, DeVries TJ, Duffield TF. A systematic review and meta-analysis of the diagnostic accuracy of point-of-care tests for the detection of hyperketonemia in dairy cows. *Prev Vet Med*, 130:18-32, 2016.
- Vande Velde F, Claerebout E, Cauberghe V, Hudders L, Van Loo H, Vercruysse J, Charlier J. Diagnosis before treatment: Identifying dairy farmers' determinants for the adoption of sustainable practices in gastrointestinal nematode control. *Vet Parasitol*, 212(3-4):308-317, 2015.
- Vang, Ó. K.,. ATP measurements for monitoring microbial drinking water quality. PhD thesis. Technical University of Denmark, Department of Environmental Engineering, 2013.
- Villot C, Martin C, Meunier B, Mialon MM, Silverberg M. Relative rumen pH thresholds to predict subacute ruminal acidosis (SARA) in dairy cows. Conference paper EcPLf, At. Nantes. <https://www.researchgate.net/publication/318225306>, 2017.
- Voyvoda H, Erdogan H. Use of a hand-held meter for detecting subclinical ketosis in dairy cows. *Res Vet Sci*, 89(3):344-351, 2010.