

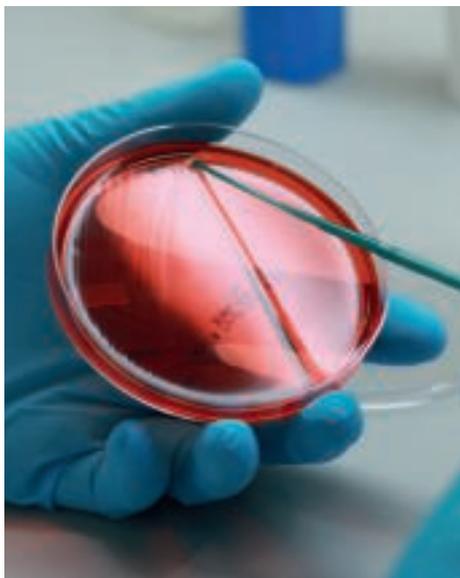
Monitoring

ANIMAL HEALTH

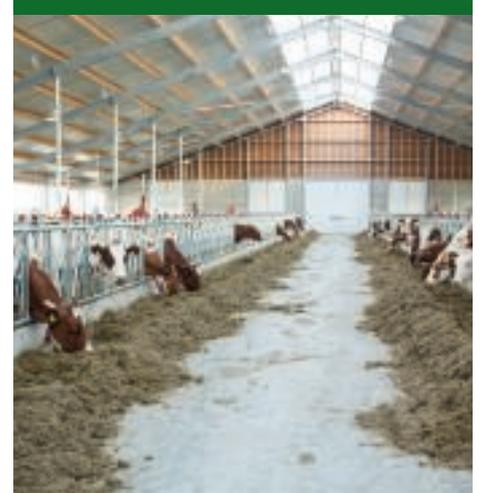


The association between antibiotic usage and resistance to antibiotics in the cattle sector

As part of the data analysis, an in-depth study determined whether there was any association between the use of groups of antibiotics on the one hand and the increased resistance of specific bacteria strains to those groups of antibiotics on the other, at the sector level, in the group of dairy farms and non-dairy farms. Over the past years, striking trends have been discernible in monitoring of the antimicrobial resistance (AMR) of pathogenic bacteria. However, it is not known whether there is a



correlation between these trends and the use of antibiotics (AMU). By linking AMU data to data on AMR, based on resistance tests conducted by GD, it became possible to analyse whether increased or decreased resistance to antibiotics is associated with changes in their use. This was done by comparing the volume of antibiotics supplied per antibiotic group with the percentage of resistant isolates, at the sector level, per quarter (dairy farms) or per annum (non-dairy farms). AMR data was available for 22 bacteria strains at dairy farms, from 2016 through 2019. Analysis required a sufficient volume of data to be available: at least 20 isolates per quarter in at least three of the four years to be analysed. Sufficient AMR data was available for seven different bacteria strains and thereby 53 pathogen-antibiotic combinations, in order to study the association between AMU and AMR, per active ingredient. A total of nine trends or significant associations were discovered between AMU and AMR, eight of which were associations with AMR of bacteria in milk from cattle suffering from (sub-) clinical mastitis. At the non-dairy farms, AMR data was studied for the nine most frequently isolated bacteria strains; an association was discovered between AMU and AMR for three types of bacteria.



This in-depth study discovered a positive association for a number of combinations of antibiotics family groups and bacteria: higher use related to a higher degree of resistance and lower use resulted in lower resistance, in the same quarter. The results of such analyses could in principle be used to draw up formularies and could therefore contribute to responsible AMU and the reduction of AMR in veterinary pathogens in the cattle sector. However, additional data is required before using the results of such an analysis. This analysis used the results of isolates from non-actuated samples, so that the results are not entirely representative of the Dutch cattle population. Retrieving more specific samples/isolates from the field would improve the representativeness of the AMR. This would increase the value of the results of such analysis and further support the responsible use of antibiotics.

Soybean meal contaminated with salmonella

A feed adviser contacted the Veekijker regarding a dairy farm which had been feeding a batch of soybean meal contaminated with salmonella in their Total Mixed Ration (TMR) over a five-day period. A soy processing factory had supplied this batch to various

farms via a fodder supplier. The contaminated batches were collected from the farms as soon as it became known that the soybean meal was contaminated with salmonella. In consultation with the feed adviser, GD reported this case to the Netherlands Food

and Consumer Product Safety Authority (NVWA). GD also advised alertness to any clinical symptoms, such as fever, diarrhoea and abortion in the cattle at the dairy farm in question, and that the attending veterinarian be informed. No salmonella outbreak occurred.

Incoming (imported) animals: risk of introducing animal diseases including paraTBC

In 2020, GD was consulted regarding the import of Jersey cattle from Denmark, whereby paraTBC was detected in faecal samples taken from a number of animals. Besides the direct damage, due to paraTBC-positive cattle having to be disposed of, longer term damage can also be suffered as the paraTBC infection can also go undetected and spread to other animals on the farm in the meantime. Incoming animals are a known risk factor for the introduction of animal diseases. Intake testing is therefore conducted in animals coming from farms with a lower or unknown status. A Danish study (Jakobsen et al., *Prev. Vet. Med.* 2000, 15-27) describes how paraTBC is most commonly found in:

- Jerseys;
- older cattle (parity more than four);
- the first month following calving;
- larger farms.

Multi-resistant *E. coli* infection in rearing calves

The Veekijker received questions about numerous rearing calves (aged 4 to 10 weeks) at a dairy farm, suffering fever, respiratory problems, neurological symptoms and swollen joints.

Based on these clinical symptoms, the decision was taken to submit two calves for pathological examination. One calf had meningitis, sepsis and polyarthritis, while the other had polyserositis and sepsis, both due to a multi-resistant *Escherichia coli*. Tests for salmonella and mycoplasma were negative.

While such diseases are known to occur in veal calves at the end of the fattening period, they

are uncommon in rearing calves. A third-choice antibiotic proved necessary to curb the outbreak and there have been no new cases since.

The advice was to thoroughly clean and disinfect the barns and drinking equipment before housing new calves. Due to the severe disease progression of the outbreak, GD bacteriologists advised storing spleen material from the two submitted calves. In the event of a repeat outbreak, this would give the option of comparing the isolated strains by means of typification, providing additional insight into the epidemiology at this farm.

Bovine tuberculosis in Belgium

In December, a case of bovine tuberculosis was reported in Belgium, via a Promed notification and DGZ news message. This concerned a cow, born in 2017, with suspected bovine tuberculosis at slaughter. PCR analysis and microscopic testing did not show any mycobacteria, but *Mycobacterium bovis* was cultivated in a bacteriological test. The cow came from a dairy farm with a herd of 278 cattle, in the Liège region. As a result of these findings, the farm was declared a source of tuberculosis. The tuberculin test was then conducted on all cattle at this farm and on cattle at 59 contact farms. Epidemiological testing is still underway. The previous source to be reported in Belgium was two years ago. At the European level, Belgium has been officially free from bovine tuberculosis since 2003. This case does not impact that status according to the Federal Agency for the Safety of the Food Chain (FAVV) in Belgium.

Summary of resistance development of bacteria at dairy farms in 2020

Throughout the year, GD reports on the resistance development of bacteria from animal material derived from dairy and non-dairy farms. This information is gained from bacteriological testing of cultures of pathogenic bacteria. A resistance test is conducted to determine the antibiotics to which these bacteria are resistant under laboratory conditions. This allows the veterinarian to make an informed choice of a particular antibiotic to treat the bacterial infection in question. The results of all the resistance tests conducted allow long-term monitoring of the development of resistance patterns among bacteria. If the number of isolates of a certain pathogen is less than 20 in any quarter, considerable restraint must be exercised when interpreting the results. In such cases, comparisons will be made once annually instead of per quarter. Only significant (P value of < 0.05) and relevant changes in antibiotic resistance are discussed here. A bacterium is defined as being multi-resistant if it is resistant to at least three different groups of antibiotics.

Pathogenic bacteria from animal material from dairy farms in 2020*

The percentage of multi-resistant isolates derived from bovine material from dairy farms in 2020 was equal to the percentage in 2019, 2017 and 2016, but significantly lower than in 2018: 43 percent in 2020 and 2019 (n=636 and n=584), 52 percent in 2018 (n=630) and 46 percent in both 2017 (n=706) and 2016 (n=670). Just like in the previous four years, of the following pathogens more than 50 percent of the isolates were multi-resistant: *Escherichia coli* F5 (=K99), *E. coli*, *Salmonella* Typhimurium and isolates typified as *Salmonella* group B (see Figure 1). The percentages of multi-resistance for individual pathogens were not significantly different in 2020 versus the percentages in 2019, with the exception of *Salmonella* Dublin, for which the percentage increased from 0 percent in 2019 (n=60) to 6 percent in 2020 (n=77). This brings the percentage back to the 2016 level (n=92; 5 percent).

* The tested isolates are derived from dead animals (isolates from necropsy material) or from sick animals (isolates from non-necropsy material), as a result of which the resistance percentages shown may not necessarily be representative of the entire Dutch cattle farming sector.

Pathogenic bacteria from milk samples in 2020

Following a significant decrease (p<0.001) in the percentage of multi-resistant isolates in 2013 (9 percent), 2014 (7 percent) and 2015 (4 percent), the percentage of pathogens from milk samples did not decrease further in 2016 through 2020. Following the trend of the previous five years, the percentage in 2020 (n=2728) is 4 percent. The percentage of multi-resistant isolates varies within the different species/groups of bacteria, from 0.1 to 9 percent (see Figure 2). No multi-resistance at all was seen in *Klebsiella* species, *Streptococcus agalactiae* and *Streptococcus dysgalactiae*. Insufficient isolates were studied for *Citrobacter* species and *Enterobacter cloacae*. The percentage of multi-resistant coagulase-negative staphylococci decreased from 12 percent in 2019 (n=362) to 7 percent in 2020 (n=247); in 2016, this percentage was also low (6 percent; n=453), before increasing for a number of years (10 percent in 2017 (n=433) and 9 percent in 2018 (n=386)). The lower percentages of multi-resistance versus isolates from materials other than milk, are related among other things to the resistance testing for a reduced number of antibiotics groups in mastitis pathogens. Isolates from samples other than milk are tested for nine to eleven different groups of antibiotics, while mastitis pathogens from milk are tested for five to six different groups of antibiotics.

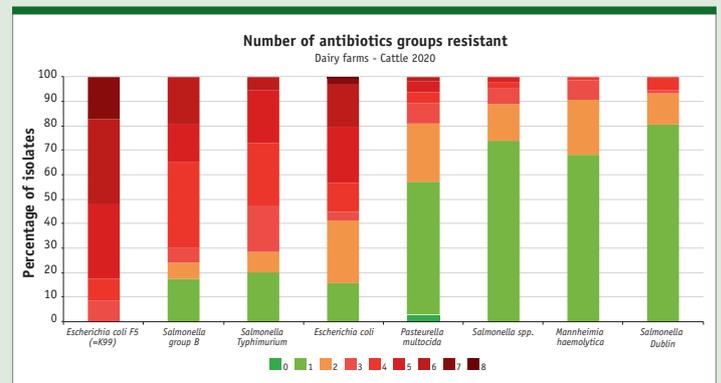


Figure 1. Per pathogen, the percentage of isolates from animal materials from dairy farms that is resistant to antibiotics of the various antibiotics groups, 2020 (0=no resistance shown, 8=resistance shown to antibiotics from eight different groups of antibiotics) (source: GD-LIMS)

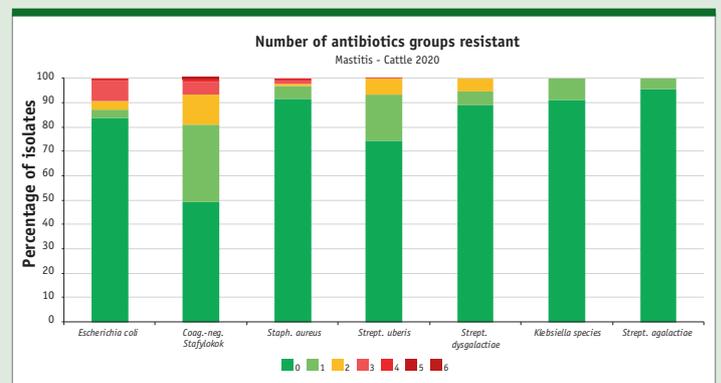


Figure 2. Per pathogen, the percentage of isolates from milk samples that is resistant to antibiotics of the various antibiotics groups, 2020 (0=no resistance shown, 6=resistance shown to antibiotics from five different groups of antibiotics) (source: GD-LIMS)

Bluetongue: situation in the Netherlands and abroad

GD conducted bluetongue screening for the Ministry of Agriculture, Nature and Food Quality in 2020. No animals with antibodies were found. The Netherlands therefore retains its BTV-free status. There were, however, notifications of cattle infected with bluetongue type-8 in France, Germany, Switzerland and Luxembourg. Luxembourg had been free from bluetongue since 2012 and has been added as a monitoring zone. A number of notifications of bluetongue type-4 were received from Eastern Europe.

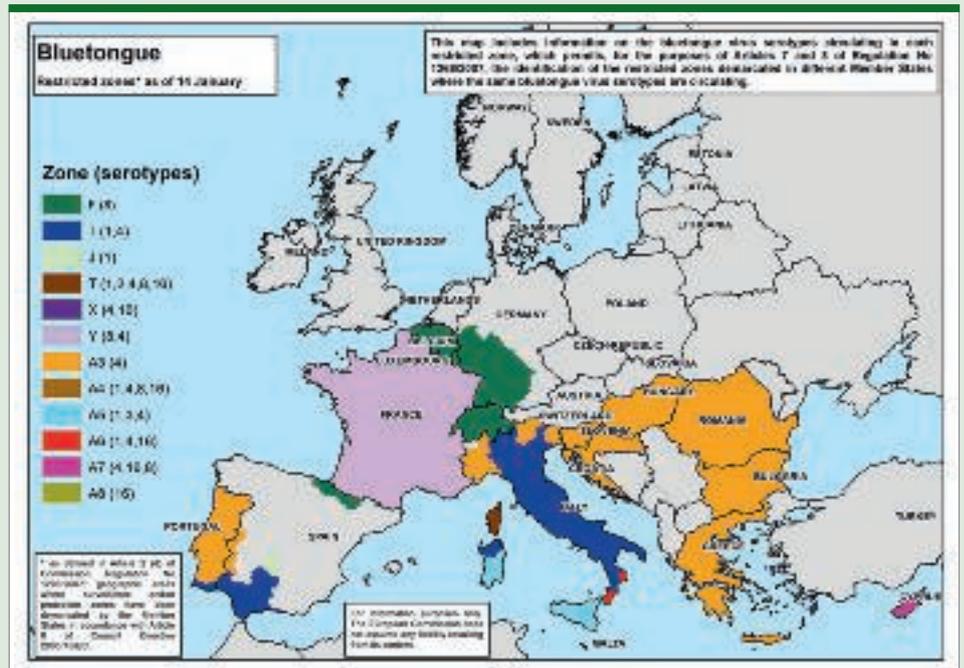


Figure 3. The monitoring zones as of 14/01/2021 and distribution per serotype of bluetongue (source: http://ec.europa.eu/food/animals/docs/ad_control-measures_bt_restrictedzones-map.jpg)

Increase in internal steatosis in cattle

Research has shown a clear increase in the percentage of cattle with internal steatosis during the period 2018 to March 2019, versus the period from 2007 to 2017. This is an undesirable situation. There is generally a strong reduction in food intake in the period around partus in cows who are overconditioned at calving. Such cows suffer a more serious and prolonged negative energy balance in the transition period and are at greater risk of associated diseases such as fatty liver, ketosis and metritis. The fatty tissue formed in internal steatosis plays a more important role than external fat (on the carcass): it produces signalling agents which results in the cow already developing an inflammatory response in the body even before calving. In order to limit the health issues suffered by cows in the transition period wherever possible, it is therefore essential to prevent steatosis in cows during the dry period. Further research is essential to discover the causes of internal steatosis in dairy cows, before determining whether practical measures should be taken. Despite a slight decline, the percentage of animals older than 1 year and submitted for necropsy with internal steatosis was still extremely high in 2020, versus the years prior to 2017 (Figure 4).

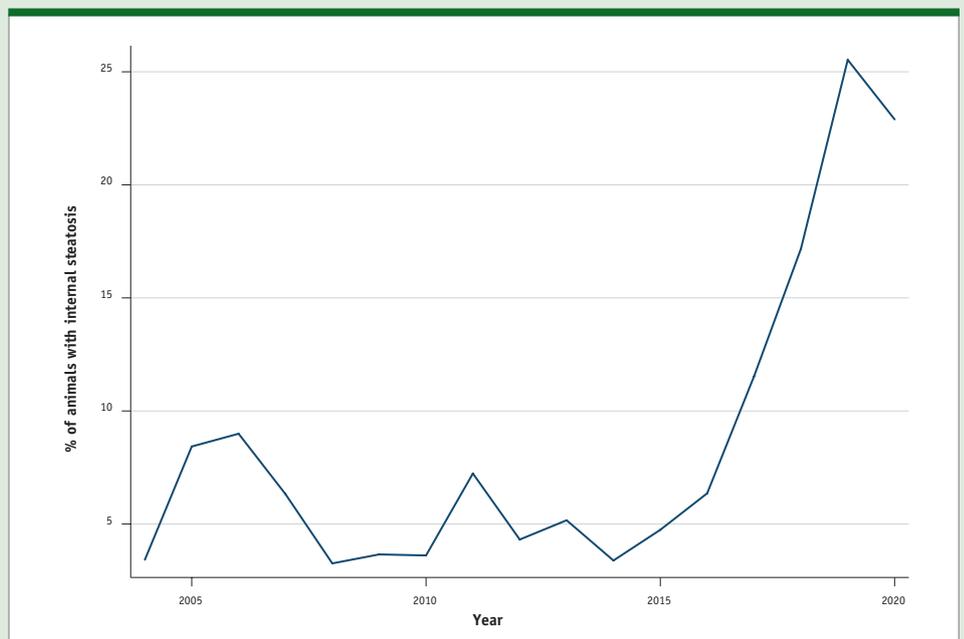


Figure 4. The annual percentage of submitted cows older than 1 year whereby steatosis was found at necropsy

Animal health barometer for cattle, fourth quarter 2020

VETERINARY DISEASES	SITUATION IN THE NETHERLANDS	Surveillance – Highlights Fourth Quarter 2020
Article 15 GWWD (Health & Welfare Act) compulsory reportable and treatable diseases (diseases named in article 2 of the 'Rules for prevention, control and monitoring of infectious animal diseases and zoonoses and TSEs')		
Bluetongue (BT)	Viral infection. The Netherlands has been officially disease-free since 2012 (all serotypes). Annual screening.	The Netherlands BTV-free, no infections detected. BTV-8 reports in Luxembourg, Germany, Switzerland and France.
Brucellosis (zoonosis, infection via animal contact or inadequately prepared food)	Bacterial infection. The Netherlands has been officially disease-free since 1999. Monitoring via antibody testing of blood samples from aborting cows.	Twelve re-tests, no infections detected.
Bovine Spongiform Encephalopathy (BSE)	Prion infection. The Netherlands has OIE status 'negligible risk'. No cases detected upon monitoring since 2010 (total 88 cases between 1997-2009).	No infections detected.
Enzootic Bovine Leucosis (EBL)	Viral infection. The Netherlands has been officially disease-free since 1999. Monitoring via antibody testing of bulk milk and blood samples of slaughtered cattle.	No infections detected.
Lumpy Skin Disease (LSD)	Viral infection. The Netherlands is officially disease-free.	Infections have never been detected.
Anthrax (zoonosis, infection via animal contact)	Bacterial infection. Not detected in the Netherlands since 1994. Monitoring via blood smears from fallen stock.	No infections detected.
Foot and Mouth Disease (FMD)	Viral infection. The Netherlands has been officially disease-free since 2001.	No infections detected.
Rabies (zoonosis, infection via bite or scratch wounds)	Viral infection. The Netherlands has been officially disease-free since 2012 (illegally imported dog).	No infections detected.
Bovine Tuberculosis (TBC) (zoonosis, infection via animal contact or inadequately prepared food)	Bacterial infection. The Netherlands has been officially disease-free since 1999. Monitoring via slaughtered cattle.	No infections detected. More infections reported in Belgium, Germany and France.
Article 100 GWWD (Health & Welfare Act) compulsory reportable diseases (diseases named in article 10 of the 'Rules for prevention, control and monitoring of infectious animal diseases and zoonoses and TSEs')		
<i>Campylobacter fetus</i> ssp. <i>venerealis</i> and <i>Trichomonas foetus</i>	Bacterial infection. The Netherlands has been disease-free since 2009. Monitoring of AI and embryo stations, and in animals for export.	No infections detected.
Leptospirosis (zoonosis, infection via animal contact or inadequately prepared food)	Bacterial infection. Control programme compulsory for dairy farms, voluntary for non-dairy farms.	One farm with antibodies in bulk milk; in total, 13 infected farms were confirmed in 2020.
Listeriosis (zoonosis, infection via inadequately prepared food)	Bacterial infection. Occasional infection detected in cattle.	Infections detected in one aborted foetus and in one cow submitted for necropsy.
Salmonellosis (zoonosis, infection via animal contact or inadequately prepared food)	Bacterial infection. Control programme compulsory for dairy farms, voluntary for non-dairy farms.	94 percent of dairy farms had favourable bulk milk results (national programme).
Yersiniosis (zoonosis, infection via animal contact or inadequately prepared food)	Bacterial infection. Detected occasionally in cattle, mostly in aborted foetuses.	No infections detected in cattle submitted for necropsy. No <i>Yersina</i> species cultivated in milk samples.
Other OIE-list diseases in the Netherlands subject to compulsory reporting		
Bovine Viral Diarrhoea (BVD)	Viral infection. Control programme compulsory for dairy farms, voluntary for non-dairy farms.	84 percent of dairy farms have BVD-free or BVD-unsuspected status. This was 17 percent among non-dairy farms.

Table continuation

VETERINARY DISEASES	SITUATION IN THE NETHERLANDS	Surveillance – Highlights Fourth Quarter 2020
Other OIE-list diseases in the Netherlands subject to compulsory reporting		
Infectious Bovine Rhinotracheitis (IBR)	Viral infection. Control programme compulsory for dairy farms, voluntary for non-dairy farms.	77 percent of dairy farms have IBR-free or IBR-unsuspected status. This was 20 percent among non-dairy farms.
Paratuberculosis	Bacterial infection. Control programme compulsory for Dutch dairy farms. 99 percent has PPN status.	78 percent of dairy farms have Paratuberculosis Programme Netherlands (PPN) status A (unsuspected).
Tick borne diseases	Vector borne diseases. Ticks infected with <i>Babesia divergens</i> , <i>Anaplasma phagocytophilia</i> and <i>Mycoplasma wenyonii</i> are present in the Netherlands.	No infections detected.
Other infectious diseases in cattle		
Malignant Catarrhal Fever (MCF)	Viral infection. Infections with Ovine herpes virus type 2 occur occasionally in the Netherlands.	One infection detected at necropsy.
Liver fluke	Parasite. Liver fluke is present in the Netherlands, particularly in wetland areas.	Infections detected at 34 farms.
Neosporosis	Parasite. An infectious cause of abortion in the Netherlands.	Infection detected in five submitted aborted fetuses.
Q fever (zoonosis, infection via dust or inadequately prepared food)	Bacterial infection. In the Netherlands, a different strain in cattle to that found in goats, with no established relationship to human illness.	One infection detected in submitted aborted fetuses.
From monitoring		
	Abomasum diseases are increasing as the main diagnosis upon pathological examination of young breeding calves.	
	Udder Cleft Dermatitis (UCD) as main pathological diagnosis continues to increase.	
	Schmallenberg detected in a calf showing congenital defects.	
Data analysis	Bovine mortality older than 1 year remains high.	
	The working life of the herd is increasing.	
	Despite a slight increase in the percentage of closed farms, the purchase of lower status animals (including import) remains a risk.	
	Udder health is declining and the use of antibiotics for mastitis is increasing.	
Resistance to antibiotics at dairy farms	No abnormalities.	
Resistance to antibiotics at non-dairy farms	No abnormalities.	

Animal health monitoring

Since 2002, Royal GD has been responsible for animal health monitoring in the Netherlands, in close collaboration with the veterinary sectors, the business community, the Ministry of Agriculture, Nature and Food Quality, vets and farmers. The information used for the surveillance programme is gathered in various ways, whereby the initiative comes in part from vets and farmers, and partly from Royal GD. This information is fully interpreted to achieve the objectives of the surveillance programme – rapid identification of health problems on the one hand and monitoring trends and developments on the other. Together, we team up for animal health, in the interests of animals, their owners and society at large.