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Changes in the Johne's disease situation in GB dairy herds over 10 years, as revealed by regular milk ELISA data

Emma Nicole Taylor[a,*,1], Kulwant Channa[a], Pete Guy Orpin[b], James Hanks[a], Nicholas Mark Taylor[c,*,2]

[a] PAN Livestock Services Ltd., School of Agriculture, Policy and Development, University of Reading, Earley Gate, PO Box 237, Reading, RG6 6EU, UK
[b] MyHealthyHerd Ltd., 10 Churchgate, Hallaton, Leicestershire LE16 8TY, UK
[c] Veterinary Epidemiology and Economics Research Unit (VEERU), School of Agriculture, Policy and Development, University of Reading, Reading, United Kingdom

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ABSTRACT

Effective management of cattle infected with Johne's Disease (JD) is crucial to minimizing transmission and within-herd prevalence. Within Great Britain (GB), the voluntary National Johne's Management Plan (NJMP) requires farmers and a certified vet to conduct a risk assessment to determine the herd risk, examine the herd JD status and formulate a management plan. Individual milk ELISA tests for JD antibodies are widely used to monitor infection. The JD Tracker application, available within the dairy data management software InterHerd+ and other web-based environments, is being used by farmers and veterinarians to facilitate the practical use of milk ELISA data to aid JD-related management decisions. The JD Tracker application uses a herd's milk ELISA data to calculate a collection of 'JD parameters' that are indicative of the current JD status of the herd alongside contemporary and retrospective drivers linked to transmission and maintenance of infection. Herein, we use milk ELISA data from 154 regularly testing herds to review the temporal trends in JD parameters from 2013 to 2022. Since 2015, JD Tracker parameters have improved in these herds, most notably average test value (ATV) and within-herd prevalence (%Pos30). Trends in driver parameters suggest that farmers are progressively less likely to serve repeat test-positive (J5) cows and are more readily removing them. The data also reveal that the burden of JD is disproportionately greater in herds with higher ATV. In 2022, the 25 % of herds with the highest ATVs accounted for 42 % of positive tests and 42 % of repeat ELISA positive (J5) cows. Retrospectively, it is not possible to identify with certainty factors that directly contributed to the trends in JD parameters, but it is notable that the introduction of the NJMP was coincided with the improving JD situation. In 2019, participation in the NJMP or an equivalent scheme became mandatory for dairy farms to be compliant with the food and farms standards assurance scheme Red Tractor, with the result that JD management plans are now completed by 95 % of UK dairy farms. As far as we know, the UK is unique in its development of a tool (the JD Tracker) which adds utility to milk ELISA data using specifically designed JD parameters. Anticipated further work includes the development of a national database of JD testing herds and application of the JD Tracker at national scale to enable more comprehensive industry-level monitoring of JD within GB dairy farms.

1. Introduction

Johne's disease (JD) is a chronic granulomatous infection of ruminants caused by *Mycobacterium avium* subspecies *paratuberculosis* (MAP). Following exposure to MAP, infected cattle enter a prolonged incubation period followed by the subclinical and clinical stages of infection (Behr and Collins, 2010). The subclinical stage is characterized by the onset of bacterial shedding within faeces (Mitchell et al., 2015), milk or colostrum (Stabel et al., 2014) with the potential transmission of MAP. The clinical stage is characterized by the onset of symptoms, including diarrhoea, weight loss and reduced milk yields, although only 10–15 % of infected cattle progress to this stage of infection (Olsen et al., 2002). The UK has an estimated JD herd prevalence of 68 %, based on bulk milk samples from 225 herds (Velasova et al., 2017) and predicted 10-year

* Corresponding authors.
E-mail addresses: emma.taylor@panveeru.net (E.N. Taylor), n.m.taylor@reading.ac.uk (N.M. Taylor).
[1] 0000–0002-8326–2799
[2] 0000–0002-6357–7259

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annual losses of ~$49 million (~£35 million) (Rasmussen et al., 2020). Other European countries have estimated lower herd prevalences of JD. The Lombardy region of Italy found a herd prevalence of 52.4 %, based on serum ELISA data from 1549 herds (Arrigoni et al., 2023). Ireland reports a herd-level true prevalence of 28 %, based on a Bayesian interpretation of individual serum ELISA data from 1039 herds (McAloon et al., 2016). Germany found herd prevalences of 3 %, 15 % and 50 % in the southern, northern and eastern regions, respectively, based on culture and IS900-qPCR data from environment samples collected from 457 herds (Eisenberg et al., 2022).

The management of JD infected cows significantly influences transmission of infection which is directly linked to within-herd prevalence. Vertical transmission from infected cow to calf is an important method of spread. Offspring of dams which are milk ELISA JD positive at the time of calving or which become MAP positive post-calving are 2.6–3.6 times more likely to become JD positive than calves of JD negative dams (Patterson et al., 2020). Monitoring of nine farms in Wisconsin demonstrated that routine milk ELISA testing and changes to heifer rearing practices, such as segregated maternity pens for ELISA-positive and ELISA-negative cattle, reduced within-herd prevalence from 11.6 % to 5.6 % over six years (Collins et al., 2010). Likewise, expert elicitation (the Delphi method) concluded that outdoor calving, limiting faecal exposure after leaving the calving house and reducing the time calves spend with their dams can reduce within-herd MAP-prevalence by approximately 37 %, 30 % and 24 %, respectively (Radia et al., 2013).

Informed by such observations, risk assessments and management plans (RAMPs) to manage infected cattle and prevent new infections are often utilised to control and reduce within-herd prevalence of JD (Sweeney et al., 2012). In Ontario, Canada, a voluntary Johne's control programme which included the completion of RAMPs ran from 2010 to 2013 (Pieper et al., 2015). The initial risk assessment produced a numerical score using 38 questions across five management areas: animal purchases; calving management; calf management; and heifer and cow cleanliness and management. Management plans were drawn up for each farm and in 2019, 180 of the 3207 farms which participated in the original control programme were recruited for follow-up risk assessments (Imada et al., 2022). Within the follow-up RAMP, farmers purchased cattle from fewer herds and the housing of sick animals in the maternity pen was less frequent. However, the overall average RAMP score increased from 126 to 144 suggesting the risk of JD transmission had increased (Imada et al., 2022). This increase in RAMP scores may suggest that farmers need more support on the implementation of JD control management plans to make the RAMP approach fully effective. In 2017, a survey of 394 UK farmers reported that 39 % experience "uncertainty on when to cull test-positive cows" and 28 – 30 % would be more likely to adopt a robust Johne's control and biosecurity plan if training for farmers was available (Orpin, 2017). RAMPs are used within the Australian (Barwell, 2023) and German (Donat and Eisenberg, 2023) national JD control programmes. Australia has not published if their control programme has impacted JD prevalence but Germany has noted decreased within-herd prevalence (Donat and Eisenberg, 2023).

In GB, with support from Dairy UK, Phase 1 of the voluntary National Johne's Management Plan (NJMP) was developed and implemented from April 2015 to December 2017, focussing on education and engagement. Phase 2, which started in January 2018 and is on-going, requires British Cattle Veterinary Association (BCVA) Johne's Certified Veterinary Advisors and farmers to conduct a risk assessment to determine the herd risk, examine the herd JD status and formulate a management plan within which farmers commit to one of six management strategies: biosecurity, protect and monitor; improved farm management (IFM); IFM and planned testing; IFM test and cull; breeding test-positive cows to terminal sire; and firebreak vaccination (Orpin et al., 2020a, b). In 2019, participation in the NJMP or an equivalent scheme became mandatory for dairy farms to be compliant with the food and farms assurance scheme, Red Tractor (Red Tractor, 2019). Since then JD management plans have been completed by 95 % of UK dairy farms (Orpin et al., 2022), with an increasing proportion of farmers selecting 'IFM and planned testing' as their control strategy (Orpin et al., 2023).

Although new diagnostic technologies are under development, including the use of bacteriophages (Swift et al., 2020), metabolomics (Taylor et al., 2022) and microRNA (Shaughnessy et al., 2020), the individual milk ELISA remains the most widely used test amongst GB farms. The National Milk Records Group (NMR) conducted approximately 1.7 million milk ELISA tests alongside routine milk recording between the beginning of 2010 and mid 2015 (Meyer et al., 2018) and testing continued to increase after 2015 (pers. comm. NMR). Milk recording organisations (MROs) use the results of tests (negative or positive) to assign a 'J-class' status to cows (from J0 to J5). MROs in UK have used a positive cut-off value of ≥30 and the J-class status depends on the test history of a cow over the sequence of tests in its lifetime. A description of how this classification system assigns J-class is given in Supplementary Table 1. Of most relevance here are: J4 which is assigned to a cow that has had its first positive result in life (90 % of J4 cases) or the first positive after a sequence of at least three negative tests that is not the first in life, and; J5 which is assigned to a cow that has had two positive results within a sequence of four. Once assigned, a cow will remain J5 for life. In addition to J-class, a further classification as 'Priority Cull' (PC) has since been added. Regardless of J-class, any cow, that has *either* two consecutive tests with result value ≥60 *or* one test with result value ≥100 is classed as PC and remains so until removed.

The JD Tracker, which was developed by the Veterinary Epidemiology and Economics Research Unit (VEERU, University of Reading) supported by the National Action Group on Johne's, was launched in July 2021 (Orpin et al., 2022). The JD Tracker application, available within the dairy data management software InterHerd+ (PAN Livestock Services Ltd.) and other web-based environments, calculates a variety of outcome measures and drivers: so-called 'JD parameters'. The outcome measures are arithmetic mean milk ELISA value (or 'average test value': ATV) and percent of milk ELISA tests that are positive (for which separate parameters are calculated using different cut-offs). The aim is to associate the outcome with other parameters to gain deeper understanding of what is driving the JD situation in a herd. The 'driver' parameters relate to persistence (% of cows categorised as J5: %J5), progression (% of cows categorised as J4: %J4), and relative risk measures related to the propensity to remove or serve J5 cows (Orpin et al., 2022). These drivers are retrospective and contemporary: because of the time interval between infection and development of detectable antibodies, %J4 and %J5 reflect disease transmission risk three or four years ago whereas relative risk of removal and service of J5 cows indicate current JD management that may influence outcome measures in the future (Orpin et al., 2022). Within the JD Tracker application, the results are presented in tabular and graphical formats designed to be easy to read. The results are colour-coded ('traffic lights') according to benchmark values based on quartiles and median values derived from a sample of herds that are already used for production parameter benchmarks. The JD Tracker presents key data enabling farmers, vets and advisors to monitor their herd's JD situation in relation to their peers, and to support informed discussion to identify areas for improvement requiring management interventions, and to register impact of such interventions. The JD Tracker calculations can be made with data from individual herds or pooled data from multiple herds, enabling progress and monitoring of an individual herd, or multiple herds within veterinary practices, milk pools or milk suppliers (Orpin et al., 2022; 2023).

The study aimed to examine the temporal trends in JD parameters and distribution of JD amongst herds. Data from 154 herds which had milk recorded and regularly used the IDEXX milk ELISA for a minimum of 10 years was used. We show improvements in these herds, most notably ATV and within-herd prevalence, document changes to the management of repeat-positive cows and highlight that the burden of JD disproportionately affects herds with high ATVs.

2. Methods

2.1. Description of the data used

VEERU and PAN Livestock Services Ltd produce an annual report on key performance indicators (KPIs) using an InterHerd+ dataset of 500 herds (PAN Livestock Services Ltd.. 2023). This study made use of the same dataset. The 500 herds were originally selected in 2010 and represented approximately 10 % of the herds in UK that routinely milk record with NMR. The criteria for selection candidates were that herds should be predominantly comprised of Holstein, Friesian or Holstein/-Friesian breeds and have had good quality monthly milk recording data for a minimum of two years. The 500 herds were selected using random numbers to ensure a representative cross-section of the herds meeting these criteria. The practice was then to retain, as far as possible, the same herds within the sample for each subsequent annual report. However, each year around 10 % of the sample either cease recording or no longer meet data quality criteria. To maintain the sample size at 500 herds, these herds are replaced by randomly selecting new herds that meet the aforementioned criteria. This study was based on the 500 herds that were present in the dataset in July 2023. Retrospective milk recording and JD testing data were available for all these herds as far back as January 2010.

For the calculation of JD parameters used in this study JD ELISA results recorded alongside routine milk recording data were used. A requirement for this study was that only JD test data from 'whole herd' tests was to be included, to exclude results from non-routine JD testing and purposively selected '30-cow screen' tests whereby only 30 cows within the herd are tested to demonstrate the existence of disease within the herd. This was achieved by identifying dates on which ELISA results were recorded for at least 50 % of cows present in a herd on that date, the 50 % threshold allowing for cows missing tests during their dry periods. The data included for each herd were results of JD tests carried out on any 'whole herd' test dates between 1st January 2010 and 31st December 2022 and the J-class status and PC status of all cows present on those dates. Subsequent service dates and exit dates of all cows present, with the time interval after the test date, were also required to calculate the relative risk measures related to removal and service of J5 cows. Data on possible services and exits was available for dates up to at least 30th June 2023, i.e. a follow-up of at least six months from the last JD test date used in the study.

Of the 500 herds in the database 472 had at least one qualifying 'whole herd' JD test between 2010 and 2022. The number of herds testing each year had risen steadily from 67 in 2010 to 421 in 2022 (51 of the 472 tested herds had stopped testing or ceased dairy farming before 2022). It was found that 323 (77 %) of the herds testing in 2022 had $\geq$1 % positive tests. Of the 421 herds testing in 2022, 209 had recorded their first 'whole herd' test before 2014, i.e. a test history spanning at least 10 years. Of these 209 herds, 55 herds had gaps of one or more years in which no 'whole herd' test was recorded: the remaining 154 herds had at least one 'whole herd' test recorded in all years from their first year up to and including 2022 (Fig. 1). In order to provide a consistent data source for examination of the temporal trends in JD parameters over the longest period it was decided to isolate these 154 herds for study. Of these 154, 48 started testing in 2010, 51 in 2011, 33 in 2012 and 22 in 2013. Since 2013 is the first year when all 154 were 'whole herd' testing, this is the year from which the trends in JD parameters are shown.

On any particular qualifying 'whole herd' test date, results of ELISA tests are recorded as the raw test value and also using a series of binary fields coded as 0/1 to denote positive results at the 'normal' cut-off (results with values $\geq$30: Pos30) and positive results with values $\geq$60 (Pos60) and $\geq$100 (Pos100). The status of all cows present on any particular qualifying 'whole herd' test date are recorded based on J-class and classification (or not) as PC. For cows tested on that date their status after taking account of the result of that day's test was used.

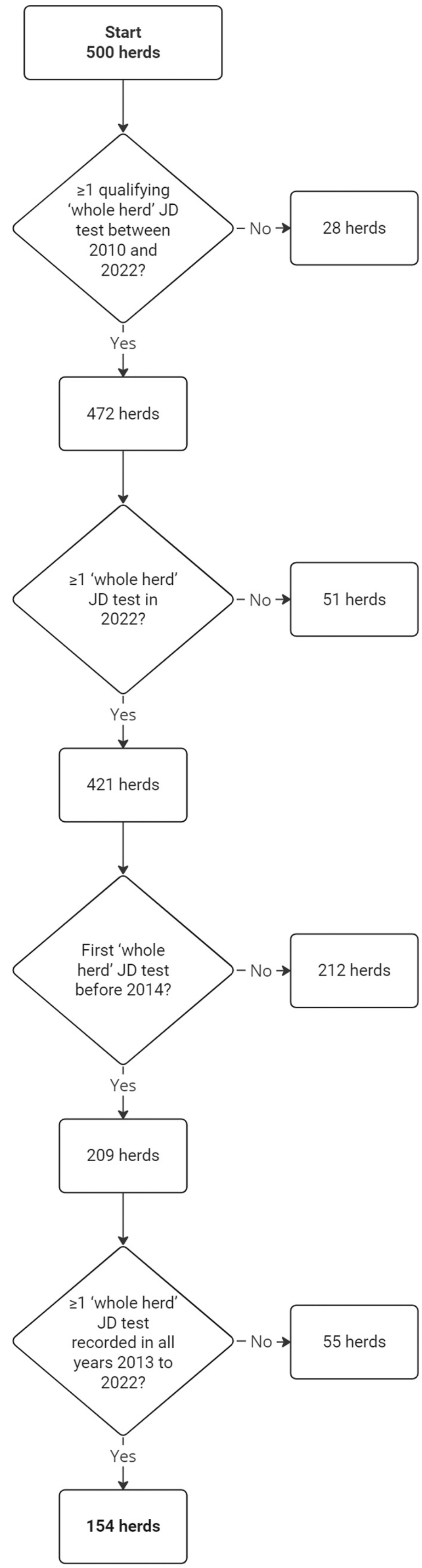

Fig. 1. Flowchart describing how and why 154 out of 500 herds were selected.

2.2. Calculation of the JD parameters

Table 1 details the definitions of the JD parameters, plus other parameters referred to in this report.

For this study annualised JD parameters were calculated at different levels of population as follows:

- annualised for each herd (by herd);
- annualised for all cows in multiple herds pooled as one population (pooled).

For cows present but not tested on a 'whole herd' test date, test result fields are "null" and not included in parameter calculations related to test results, such as ATV or %POSx. For example, for a herd with 100 cows in which an average of 80 % of the cows are tested four times a year, the annualised parameter %Pos30 would be the total number of positive tests (≥30) recorded from all 'whole herd' test dates in a calendar year divided by the total number of tests carried out (i.e. 320). Parameters related to the J-class of cows, such as %J5, are calculated using data from all cows present on the test dates. For the same herd as in the example above, the annualised parameter %J5 would be the sum of classifications of cows as J5 on all 'whole herd' test dates in a calendar year divided by the sum of cows recorded as present on all those dates (i.e. 400). Note that the same cow present on all four test dates and classified as J4 on one test and J5 on the other three test dates would contribute '3' to the numerator and '4' to the denominator of the annualised %J5 parameter (reasonably implying that cow was J5 for ¾ of the year).

The same methods apply to calculation of parameters for pooled populations, the only difference being that all data are pooled and summed across all herds included, as if one 'global' herd.

The parameters are produced by a single continuous procedure coded in SQL and placed in an output grid. The query could be set to output annualised parameters for each herd for each calendar year or aggregate parameters for multiple herds for calendar years. The output grids were exported to an Excel workbook (Microsoft Excel 2016) and Statistix version 10 (Analytical Software, Tallahassee, USA) where summary statistics, including quartiles and medians could be calculated.

Table 1
Definition / description of ANNUALISED JD parameters.

nCows	Sum of counts of cows present in herds on 'whole herd' test dates in a calendar year e.g. in a 100 cow herd with four 'whole herd' test dates the sum of counts would be 400
nTests	Sum of JD tests with valid results recorded on 'whole herd' test dates in a calendar year
%Test	Percent of cows present that are tested on 'whole herd' test dates = nTests / nCows
AveTVal	Arithmetic mean of test result values (of all tests in a calendar year) = sum of test values recorded / nTests
%Pos30	Percent of positive tests (≥30) among all test carried out = sum of tests with value ≥30 / nTests
%Pos60	Percent of test results ≥60 among all test carried out = sum of tests with value ≥60 / nTests
%Pos100	Percent of test results ≥100 among all test carried out = sum of tests with value ≥100 / nTests
%J4	Percent of J4 cows among all cows present = number of J4 classifications recorded over a calendar year / nCows
%J5	Percent of J5 cows among all cows present = number of J5 classifications recorded over a calendar year / nCows
%PrCulls	Percent of 'Priority Cull' cows among all cows present = number of Priority Cull classifications recorded over a calendar year / nCows
RRservJ5	Ratio of proportion of J5 cows with a subsequent service compared with proportion of cows with J-class status 'unclassified', J0, J1, J2 or J3 (i.e. ALL except J4 and J5) served
RRexitJ5	Ratio of cull risk within 150 days after a test date for J5 cows compared with cull risk for cows with status 'unclassified', J0, J1, J2 or J3 (i.e. ALL except J4 and J5)

2.3. Distribution of JD burden among herds in 2022

Herds were split into quartiles based on their annual ATV for 2022. Using the raw JD test data the total numbers of positive JD tests (at different cut-offs) and total numbers of J5 and PC cows to be found in the herds in each ATV quartile were calculated. The relative distributions of these numbers by ATV quartile were derived for graphic representation.

Also, using the total numbers of JD tests and total numbers of cows present in the herds in each ATV quartile, the percentages of positive tests, J5 and PC cows in each ATV quartile were calculated. To provide a 'real-life' illustration of the comparative JD burdens in herds within each quartile, these percentages were applied to a herd with the overall average size (of all herds in the dataset). This illustration is presented as a graphic detailing the average expected numbers of cows with positive JD tests and classified as J5 or PC in comparable 'typical' herds within each ATV quartile.

3. Results

3.1. Overview of 'whole herd' testing history

Supplementary Fig. 1 shows the numbers of JD tests recorded each year in the 154 studied herds and in the other (excluded) herds (note that only tests carried out as part of 'whole herd' tests are counted). The number of tests in the 154 studied herds increased from 23,287 in 2010 to 103,693 in 2014, and since then the number increased slightly to 131,756 in 2022. The 154 studied herds accounted for around 85 % of all tests in the years from 2010 to 2014 inclusive, after which testing in the other herds began to increase, from 18,531 in 2014 to around 130,000 in 2021 and 2022.

Since 2013, 96 (62.3 %) to 125 (81.2 %) of the 154 studied herds had four or more 'whole herd' tests per year (annual average 3.4–3.9 tests per herd). Notably, since 2014, at least 75 % of the 154 studied herds had four or more 'whole herd' tests per year (Supplementary Fig. 2).

We compared the annual 'pooled herds' ATV for the 154 studied herds, for the other (excluded) herds and for all herds pooled together, as shown in Supplementary Fig. 3. The ATV was similar for all three pools from 2016 onwards. In the years before 2016, the ATV for excluded herds differed from the studied herds, but since these contributed 15 % or fewer tests to the annual total until 2015 the ATV for the studied herds did not differ from the ATV for all herds. This indicates that the JD status and the change in disease situation over time are not materially different in the 154 herds selected for detailed study compared with the herds excluded from the detailed study.

On average, between 81.2 % and 84.4 % of cows that were present on the 'whole herd' test dates in any year were actually tested on those dates (Supplementary Table 2). It should be noted that this does not mean that between 15.6 % and 18.8 % of cows were never tested in any particular year, because when a herd is testing more than once per year different cows will miss tests on each occasion, usually as a result of being 'dry' so not accessible for milk sampling. In fact, with multiple chances to be tested each year, the aim is that all cows should be tested at least once a year

3.2. Temporal trends in JD parameters from 2013 to 2022

Figs. 2 to 5 show the JD parameters calculated for the 154 studied herds as one 'pooled herd' by calendar year, therefore highlighting the change in each parameter over the years 2013–2022 across the cow population within the herds as a whole. Fig. 2 shows the ATV of all JD tests carried out during 'whole herd' tests in the 154 herds, each year. For reference, the timelines of the development of the NJMP phases and the JD Tracker are also indicated over the chart.

Overall, ATV has decreased from 8.44 in 2013 to 6.21 in 2022, having initially increased to 12.3 in 2015. Since 2015, ATV trended downwards, with the exception of a small increase to 9.05 between 2018

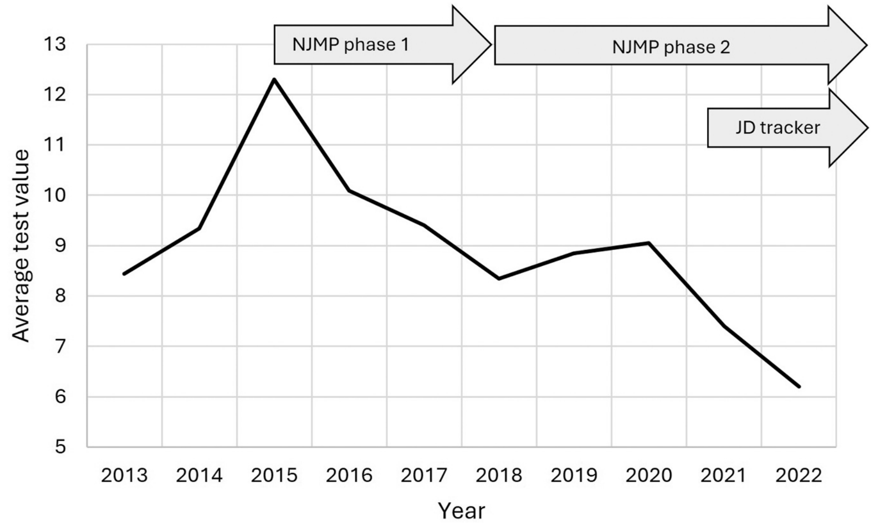

Fig. 2. The pooled ATV for JD tests carried out during ‘whole herd’ tests in the 154 studied herds, by year. NJMP Phase 1 and 2 refers to the National Johne’s Management Plan Phases 1 and 2.

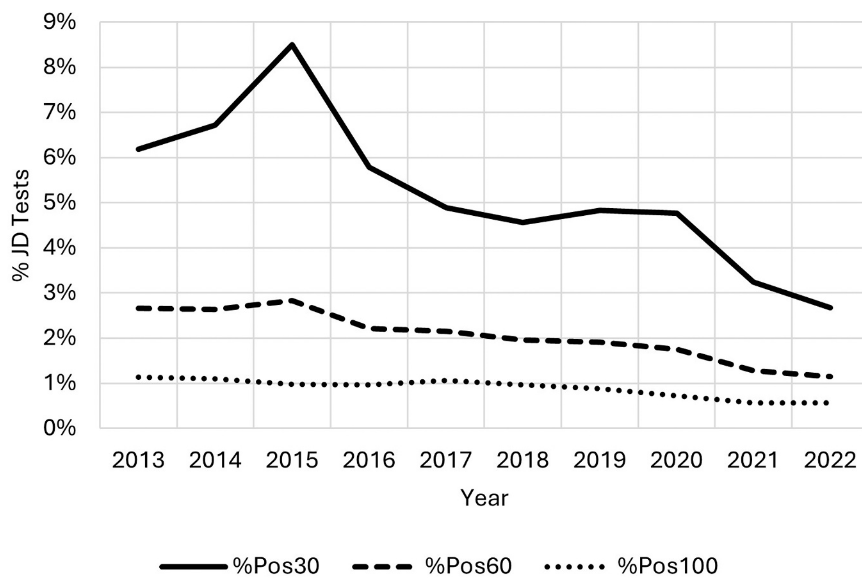

Fig. 3. The pooled % positive, at different cut-offs, for JD tests carried out during ‘whole herd’ tests in the 154 studied herds, by year.

and 2020 (Fig. 2). The percent of positive JD tests, as indicated by %Pos30, demonstrated the same trend as ATV between 2013 and 2022. Overall, %Pos30 more than halved, decreasing from 6.19 % in 2013 to 2.68 % in 2022. There was an initial increase to 8.49 % in 2015, but %Pos30 then decreased, with the exception of small temporary increases in 2019 (4.83 %) and 2020 (4.76 %) (Fig. 3).

Interestingly, a much smaller peak was observed in %Pos60 in 2015 compared with that in %Pos30 and there was no such feature in %Pos100. %Pos60 more than halved from 2.66 % in 2013 to 1.14 % in 2022, after a small increase to 2.83 % in 2015. Having fluctuated between 1.10 % and 0.88 % for six years between 2014 and 2019, %Pos100 finally more than halved from 1.13 % in 2013 to 0.56 % in 2022 (Fig. 3).

The percent of cows being categorised as a J4, J5 or PC also more than halved between 2013 and 2022: %J4 from 2.80 % to 1.33 %; %J5 from 4.75 % to 2.24 %; %PC from 2.21 % to 1.02 %. The %J4 and %J5 parameters also demonstrated a notable peak in 2015, followed by smaller peaks in 2019/2020 (Fig. 4).

Fig. 5 shows the relative risk of subsequent service and relative risk of exit within 150 days after ‘whole herd’ test date, comparing risk in J5 cows with all other cows except J4 cows, for the pooled herd populations by year. The relative risk of serving a J5 cow has decreased overall from 0.75 in 2013 to 0.40 in 2022. The relative risk of a J5 cow exiting the herd has increased from 2.29 in 2013 to 3.09 in 2022.

3.3. Changes in distribution of parameter values among herds

Fig. 6 contains box and whisker plots showing the distribution of annualised JD parameters by herd, among the 154 studied herds. In these plots the rectangles cover the middle 50 % of herds, with the position of the median marked by a horizontal line. The whiskers extend below and above the box to indicate the minimum and maximum values, excepting any outliers that are shown as individual points. The plots were created in Excel which considers any data value to be an ‘outlier’ if it is 1.5 times the interquartile range (IQR) larger than the third quartile or smaller than the first quartile. Note that because the vertical axes on the plots have been scaled to focus on the box and whiskers, some outliers are higher than the axis maximum and therefore not visible (off-

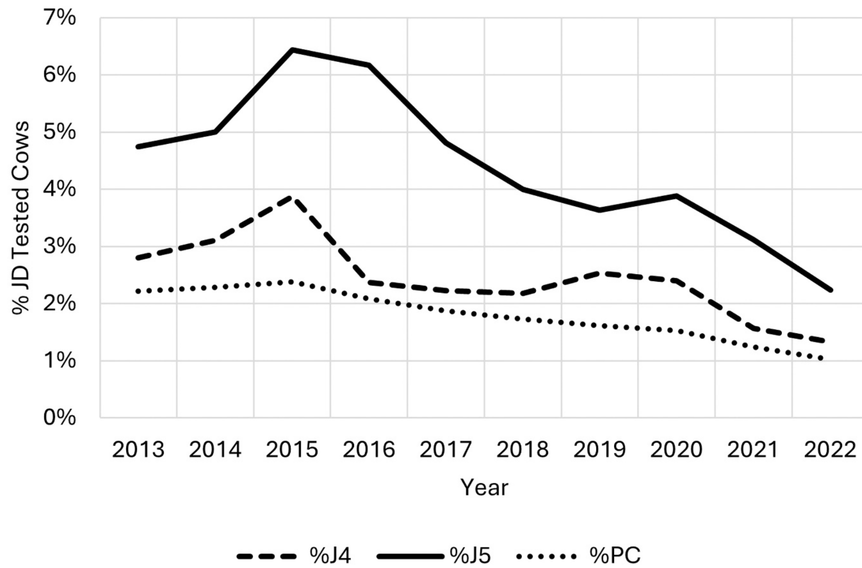

Fig. 4. The pooled % J4, J5 and 'PC' classified cows among cows present on 'whole herd' test dates in the 154 studied herds, by year.

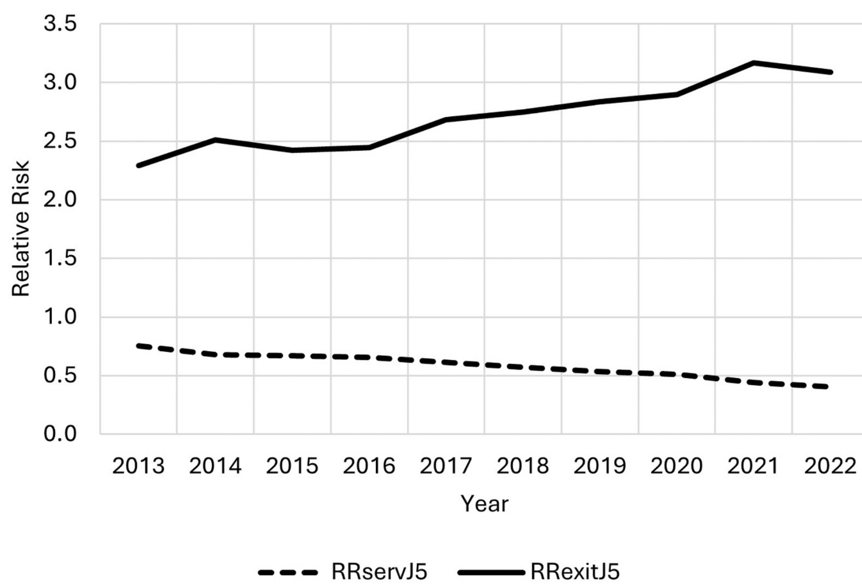

Fig. 5. The pooled RR of subsequent service and RR of exit within 150 days after 'whole herd' test date comparing risk in J5 cows with all other cows except J4 cows present on test dates in the 154 studied herds, by year.

scale).

3.4. Distribution of JD burden among herds in 2022

The 154 studied herds were split into quartiles based on their annual ATV for 2022. In fact this gave a split of 38, 39, 39 and 38 herds in quartiles 1, 2, 3 and 4. The average herd size in the four quartiles also differed, being 225, 293, 309 and 250 in quartiles 1, 2, 3 and 4 respectively, with an overall average herd size of 270. Therefore the 38 herds in quartile 1, with the lowest ATVs, contained 21 % of the cows; quartile 2, 29 %; quartile 3, 28 % and quartile 4, 23 % (Fig. 7). The relative distribution of positive JD tests (at different cut-offs) and total numbers of J5 and PC cows by ATV quartile are shown in Fig. 7.

In these 154 herds, 42.1 % of all JD positive tests originated from herds in quartile 4, with the highest ATVs. The 50 % of herds in quartiles 3 and 4 together accounted for 73.8 % of all JD positive tests. Similarly, 44.7 % and 46.6 % of all Pos60 and Pos100 JD tests originated from herds located in quartile 4 and 78.4 % and 81.5 % of all Pos60 and Pos100 JD tests originated from the 50 % of herds with the higher ATVs. With respect to cow categories, 41.9 % of J5 cows and 44.6 % of PC cows were located in herds in quartile 4 and 74.1 % of J5 cows and 78.5 % of PC cows were located in the 50 % of herds with the higher ATVs. In contrast, just 5.3 %, 4.1 % and 3.1 % of the Pos30, Pos60 and Pos100 JD tests, and 5.8 % and 3.9 % of all J5 and PC cows were attributable to herds in quartile 1 (Fig. 7).

Fig. 8 provides a 'real-life' illustration of the comparative JD burdens in herds within each ATV quartile, where the actual percentages of positive tests, J5 and PC cows were applied to 'typical' comparable herds with the same overall average size (270 cows). Herds in quartile 4 typically accounted for 7.6 more ELISA positive tests, 6.5 more J5 cows and 10.8 more priority cull cow than herds in quartile 1.

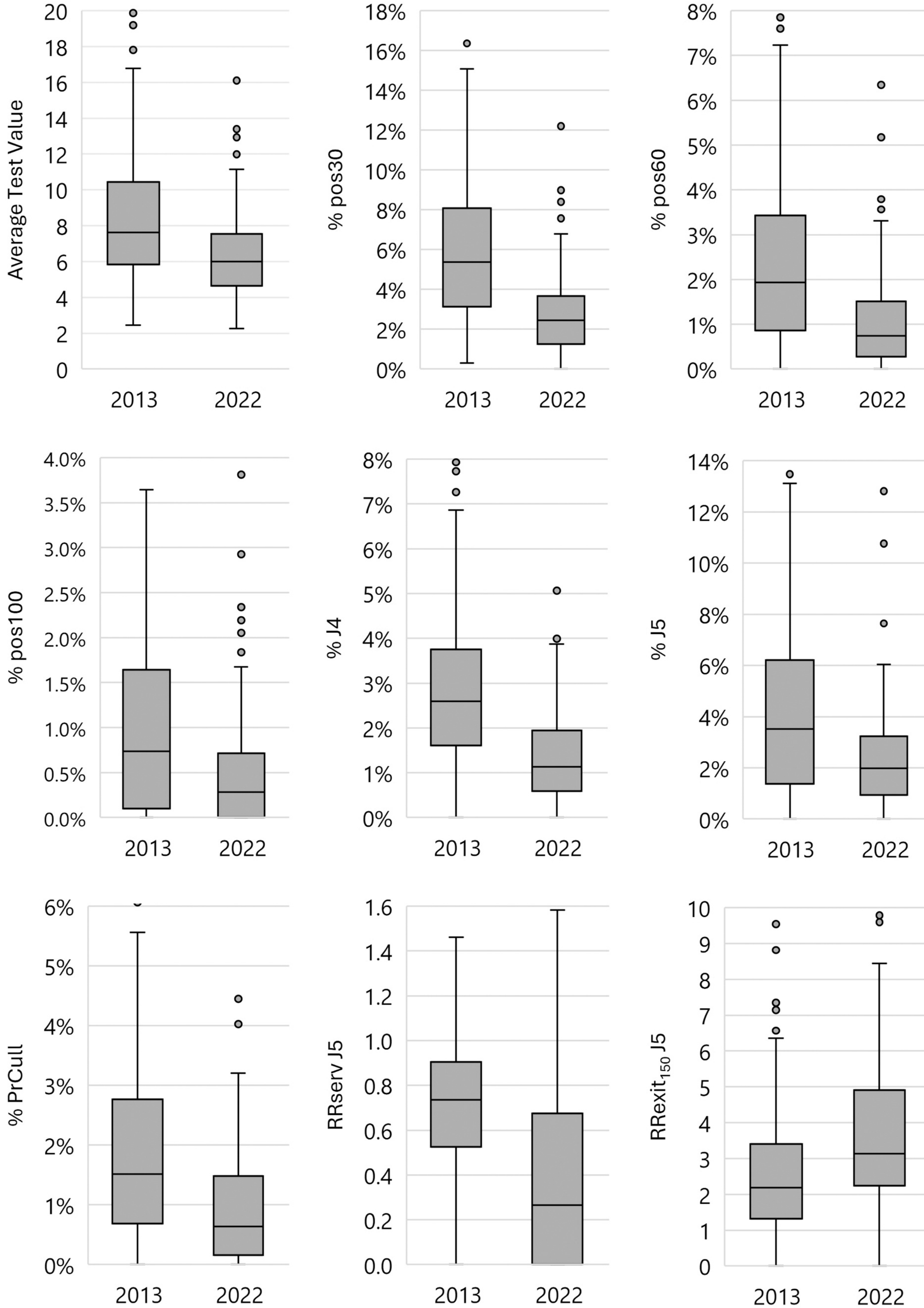

Fig. 6. Box and whisker plots of herd annualised JD parameters in 154 studied herds, comparing 2013 with 2022.

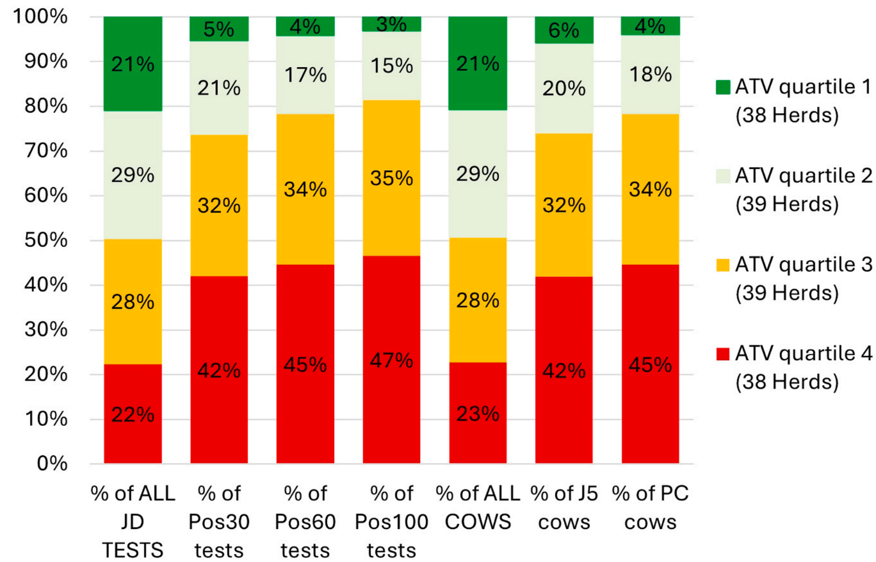

Fig. 7. Relative distribution of absolute numbers of positive tests and J5 and PC cows found in the 154 studied herds in 2022, by herds in each ATV quartile in 2022.

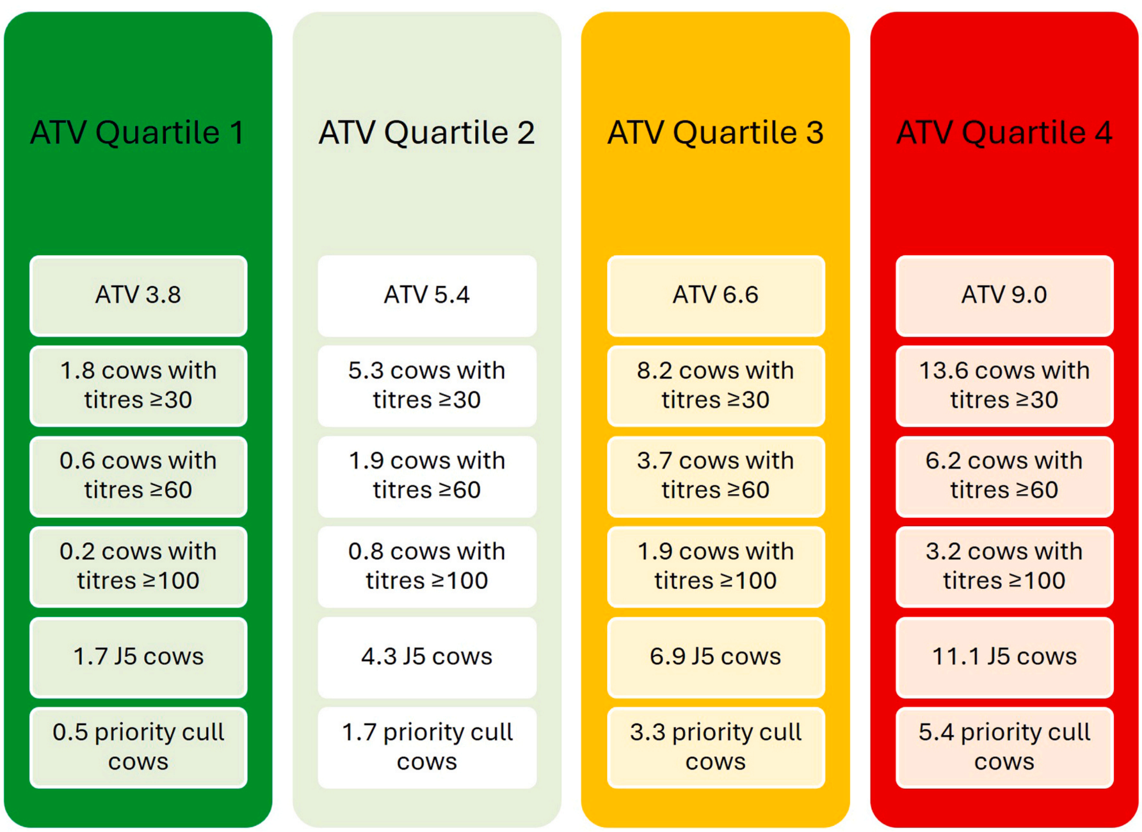

Fig. 8. Herd profiles detailing the overall ATV, average expected number of cows with milk ELISA titres >=30, >=60, >=100 and number of J5 and PC cows to be found in a typical 270 cow herd within the best quartile (1), quartile 2, quartile 3 and worst quartile (4) for ATV (2022 data).

4. Discussion

Data from a consistent dataset of dairy herds has been used to generate benchmarks for KPIs since 2010 (PAN Livestock Services Ltd., 2023) but analysis of milk ELISA results from these herds was only included in annual reports since 2018 and has been *ad hoc*. Other literature on long-term trends in milk ELISA data is limited. Identifying trends in consistent and comparable JD Tracker parameters will allow industry, particularly Action Group Johne's (an open forum for industry stakeholders interested in tackling Johne's disease) and commercial retailers, to determine the impact of JD control and management approaches and to make more informed policy decisions. Milk recording and milk ELISA data from 154 herds which had been 'whole herd' testing for a minimum of 10 years was used to determine trends in JD Tracker parameters between 2013 and 2022. The data from these herds for 2022 was also used to examine the distribution of JD burden within these herds. The 154 studied herds can be considered representative of the population of GB Holstein/Friesian dairy herds that routinely milk-record and regularly use JD testing, in respect to herd size and production characteristics. In support of this is the fact that these herds were drawn from a random selection of 500 herds that has long been accepted as representative in terms of production parameters, including herd size, production, fertility and health. However, it should be pointed out that the subset of 154 studied herds were purposely chosen on the basis of having 'whole herd' JD tests in every year since 2013 or earlier. All the managers of these herds have chosen to regularly test for JD voluntarily. This is presumably because they have concern about the prevalence of JD in their herds, with some at least engaging in JD control activities. Therefore the picture presented by this study of JD level and changes over time may apply only to such herds and not to non-testing herds. It should also be noted that this study cannot be used to estimate between herd prevalence of JD in GB, although this is likely to be high, since 421 herds of the 500 herd dataset had carried out a 'whole herd' JD test in 2022 and 323 (77 %) of these had ≥1 % positive tests.

4.1. JD prevalence and observed trends

In 2022, the 154 studied herds demonstrated an overall individual prevalence (% of tests positive) of 2.68 % (Fig. 3) with the median herd having a within-herd prevalence of 2.46 % (Fig. 6). This prevalence falls within the range found in other European countries. Italy demonstrates a lower animal-level JD prevalence of 1.3 %, based on serum ELISA data from 173,034 cows (Arrigoni et al., 2023). Meanwhile, researchers in Ireland estimated a higher median within-herd true prevalence of 3.2 %, based on a Bayesian interpretation of individual serum ELISA data from 1039 herds (McAloon et al., 2016). However, the use of different survey protocols and diagnostic methods to determine the JD prevalence's across Europe make direct comparisons problematic because these surveys would have different sensitivities and specificities (Nielsen and Toft, 2008).

The parameter %Pos30 used in this analysis is comparable with measures of within-herd sero-prevalence reported elsewhere. However, as a result of the development of the JD Tracker, herd ATV is being widely adopted in GB as a key indicator of JD level in the herd (Orpin et al., 2022). The herd ATV has been shown to be strongly correlated with %Pos30 (Orpin et al., 2022) but ATV is a more nuanced parameter because rather than using a binary 'pos/neg' measure it takes full account of test values just below the 30 cut-off (cows that may be in the early stage of infection) and is also 'weighted' by cows with very high test values. These high test value cows are rightly prioritised for attention because they contribute disproportionately to the herd 'JD load' and have particular significance in disease transmission (Orpin et al., 2022).

In studied herds, the overall changes in herd-level JD parameters between 2013 and 2022 are best visualised by comparing the box and whisker plots for 2013 and 2022 (Fig. 6). These not only show the change in median herd values, but also show the change in spread of values among the herds. With the exception of management parameters (RRServJ5 and RRExitJ5), improvements were observed in all quartile values, but the Q3 values made relatively bigger improvements (reductions) than the lower Q1 values, thus narrowing the IQR so that the herds demonstrated less variation in 2022 compared to 2013. For example, the IQR for %Pos30 narrowed from 5.0 in 2013 (3.1–8.1 %) to 2.4 in 2022 (1.3–3.7 %). In contrast, the IQR for RRservJ5 increased from 0.38 (0.53–0.90) to 0.67 (0.00–0.67) and the IQR for RRexitJ5 increased from 2.08 (1.32–3.40) to 2.66 (2.24–4.90) indicating that the more proactive herds improved their management of J5 cows faster than less engaged herds. While the median herd ATV has reduced by a relatively small amount (1.6 units) the values for Q3 and the maximum (excluding outliers) have both reduced by more, Q3 by 2.9 units and the maximum (excluding outliers) by about 5.6 units. This shows that the JD situation is improving across all studied herds and furthermore, those herds that started from a worse baseline (higher ATV) have tended to improve faster. Despite this, as the 'better' herds have also improved, the 50 % of herds with the higher ATV in 2022 still carry a disproportionate share of the JD burden (Figs. 7 and 8).

The long-term trends in the pooled ATV and %Pos at all three cut-offs (≥30, ≥60 and ≥100) were downwards in the studied herds, with %Pos measures more than halved from 2013 to 2022 (Figs. 2 and 3). In the ATV and %Pos30 charts 2015 appears as a 'spike', with distinctly higher values than 2014 and 2016 (Figs. 2 and 3). In comparison, there is only a slight increase in %Pos60 in 2015 compared with 2014 and 2016, and there is no visible spike in %Pos100 in 2015 (Fig. 3). This indicates that the observed spike in ATV and %Pos30 was caused by an increase in the number of tests with values ≥30 but <60. The spike in %Pos30 is reflected in the temporal pattern for %J4 (Fig. 4), since J4 classification occurs when a cow experiences its first positive result (or first after a sequence of at least three negatives). The spike in %Pos30 also leads to the peak in %J5 over the same period (Fig. 4). As cows are categorised as J4 after one positive JD test but cannot be categorised as J5 unless they have demonstrated repeated positive JD tests, %J5 may increase later than any increase in %J4. For example, %J4 increased between 2018 and 2019, but a corresponding increase in %J5 was not observed until 2020. It should be noted that not all J4 cows necessarily progress to being categorised as J5 cows and J5 cows may be subject to targeted culling, so the changes in %J5 are not wholly and exclusively dependent on changes in %J4 (Fig. 4).

The reasons for the 2015 spike in the number of tests with values ≥30 but <60 remains unclear. It is known that the milk ELISA for JD has increased sensitivity but decreased specificity if used within 28 days after the single intradermal comparative cervical tuberculin test (SICCT) for bovine tuberculosis (bTB) (Nunney et al., 2022). Therefore, use of the milk ELISA for JD shortly after the SICCT could cause a peak in some JD parameters in an individual herd. However, it seems unlikely that a large enough proportion of the studied herds would have scheduled JD testing shortly after the SICCT in a single year (i.e. 2015) sufficient to cause a spike in JD positive tests noticeable at multi-herd level. Examination of bTB data collated by the Animal and Plant Health Agency (2023) showed that the annual number of bTB tests conducted and incidents in England and Wales gradually increased before and after 2015, but there were no rapid increases or decreases. More relevant data on the timings between the use of the bTB SICCT and JD milk ELISA are unavailable for individual herd tests.

The JD Tracker management parameters do not indicate that the management of J5 cows in the studied herds could have been linked directly with this short-term spike, but are consistent with the overall improvement in JD prevalence parameters. The RR of serving a J5 cow has decreased and the RR of a J5 cow exiting the herd increased from 2013 to 2022, indicating that farmers are relatively more frequently deciding to remove J5 cows from the herd instead of breeding from them again. The probability of serving a J5 cow was lower than that for other cows (RR<1) throughout the period and the RR has reduced consistently since 2013 (Fig. 5). A more marked reduction in RR for J5 service has occurred for the 'best' quartile of herds (Fig. 6), approaching zero from 2021 (i.e. hardly any probability to serve a J5 cow) (Fig. 5). Similarly, the probability of removing a J5 cow was higher than that for other cows (RR>1) throughout the period (at least twice the probability) (Fig. 5). The median herd RR for J5 exit has increased since 2013, with Q3 going above two in 2022 (Fig. 6).

4.2. Engaging dairy farmers in JD control

In line with the findings of this study, many European countries have reported decreasing JD prevalence in the past decade alongside engagement with control strategies. Following updated national guidelines issued by the Italian Ministry of Health, the proportion of high risk herds (within-herd apparent prevalence >5 %) in the Lombardy region of Italy reduced from 28.8 % in 2018 to 4.2 % in 2021, based on individual serum ELISA data from up to 1549 herds (Arrigoni et al., 2023). In this case the fastest reductions in the proportion of high risk herds were apparent from 2017, four years after introduction of the new guidelines (Arrigoni et al., 2023). Meanwhile, in the Tyrol province of Austria, the between herd prevalence reduced from 7.5 % in 2013 to 0.97 % in 2016/17 and 0.5 % in 2019, based on boots swab culture and PCR data from 4000 herds. Boot swabs were not collected in 2014 or 2015, so the precise timing of reductions in JD prevalence is unclear (Khol, 2023).

JD control programmes within developed, endemically JD infected countries demonstrate shared aims to improve biosecurity and minimize prevalence, but funding sources, control activities and diagnostic testing requirements vary widely (Geraghty et al., 2014). The main area of contrast between GB and other countries' control programmes are the diagnostic testing requirements. Within Austria and Germany testing protocols are focused on a pooled test (e.g. using a sample from the bulk milk tank), with follow-up individual faecal sampling or milk ELISA testing, respectively (Khol, 2023; Donat and Eisenberg, 2023). Within Italy and the Netherlands, testing protocols are exclusively focused on individual milk or serum ELISA testing (Arrigoni et al., 2023; Weber et al., 2023). Furthermore, in Australia, Italy and the Netherlands, herds are classified according to the results of these diagnostic tests, for example, in Australia herds are given a score between 0 (the worst) and 8 (the best) (Barwell, 2023).

Within the NJMP in GB, participating farmers are required to commit to one of six control strategies as described in the introduction of this paper, but diagnostic testing is not a compulsory component of all six options (Orpin *et al.,* 2020). Large-scale, long-term monitoring of the JD prevalence has been historically challenging, but the use of planned 'whole herd' testing is increasing in popularity with wider engagement in the NJMP (Orpin et al., 2023).

As far as we know, the UK is unique in its development of a tool which adds utility to milk ELISA data using specifically designed JD parameters. The JD Tracker was developed to maximise the decision support value of the increasing volume of JD test data. The effectiveness of the JD Tracker in monitoring JD prevalence and identifying management areas for improvement has been demonstrated in case studies: for example, an individual herd improved their within-herd prevalence from 29.7 % to 3.0 % in five years. Over the same time period, 23 JD testing herds covered by the linked veterinary practice improved their average within-herd prevalence from 9.6 % to 4.0 % (Orpin et al., 2022). It is recognised that this study does not provide evidence of a 'cause-and-effect' link between development of the NJMP and improvement in the JD parameters since 2015. However, such improvements occurred against a background of implementation of a suite of JD control measures (which included the JD Tracker tool) with participation in the NJMP or an equivalent scheme becoming mandatory for dairy farms to be compliant with the Red Tractor Assurance scheme since 2019. Although milk ELISA testing has been available and used since 2010, it is an epidemiological paradigm that testing alone does not control disease. The observed improvement in disease level among these 154 regularly testing herds was contemporary with the introduction of clear control interventions under the NJMP and latterly with the support of enhanced analysis and farmer-targeted presentation of the testing data using the JD Tracker (Fig. 2), and it is not unreasonable to suggest that these interventions could have aided the observed improvement.

Analysis carried out in this study has shown that the burden of JD is disproportionately greater in the herds that have the higher ATVs. A typical herd in the worst quartile, with an overall ATV of 9.0 in 2022, has 7.5 times as many cows with positive JD tests ≥30, 10.3 times as many cows with positive JD test ≥60 and 16 times as many cows with positive JD tests ≥100, compared to a typical herd in the best quartile with an overall ATV of 3.8 in 2022. Likewise, a typical herd in the worst quartile for ATV has 6.5 times as many J5 cows and 10.8 times as many PC cows compared to a typical herd in the best quartile (Fig. 8). These figures have proved very useful in meetings with stakeholders in the GB dairy industry to show that while many herds have improved greatly, the herds with the higher ATV contribute disproportionately to the disease load. The message to be taken from this is that any national control plan will need to pay particular attention to herds that fall behind in the struggle to reduce infection.

A series of in-depth interviews with farmers with mixed levels of engagement with JD control and veterinarians examined the management of JD in the UK. These reported four main challenges: space; managing farmer expectations; 'free rider' issues where farms with a high prevalence of JD can still participate in the NJMP or equivalent schemes, without showing improvement in herd prevalence and continue to sell cattle without the purchaser being aware of their poor JD performance; and challenges in vet-farmer communication (Morrison et al., 2023). Moreover, Orpin et al. (2023) suggested that the NJMP could be strengthened by standardised risk assessments and methods to measure within-herd prevalence.

Going forwards, the policy of milk buyers is likely to strongly influence the uptake of planned testing using the milk ELISA on an annual or quarterly whole-herd test basis. A national JD Tracker based on a database of JD test data from as far as possible all regularly testing herds would enable the development of regularly updated quartile and other statistics for JD parameters. Such parameters, fully anonymized and centrally captured would provide universal reference points for use by MROs.

5. Conclusions

Over the last 10 years, a subset of 154 herds has experienced considerable improvements in JD Tracker parameters, particularly ATV and within-herd prevalence. Over the same period, farmers' management of cows most likely to be infected has changed: farmers are less likely to serve repeat ELISA positive cows and are more readily removing them. However, the burden of JD remains disproportionately greater in the herds with the higher ATV.

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Emma Nicole Taylor: Writing – review & editing, Writing – original draft, Visualization, Project administration, Methodology, Investigation, Formal analysis. **Peter Guy Orpin:** Writing – review & editing, Supervision, Conceptualization. **Kulwant Channa:** Software, Resources, Data curation. **Nick Mark Taylor:** Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Methodology, Investigation, Formal analysis, Conceptualization. **James Hanks:** Writing – review & editing, Supervision, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.prevetmed.2024.106317.

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