Spring Surveillance Programme Specifications

1. A spreadsheet detailing the scheduled base sampling and a flow diagram representing the full sampling plan are provided at the conclusion of this document (appendix 1 and 2).

2. BTM samples will be collected every 2 weeks commencing 4 weeks after the commencement of lactation. A total of 6 samples will be collected (week 4, 6, 8, 10, 12 & 14 post planned start of calving (PSC)). Commencement of lactation will be defined as the date that the 1st sample of BTM is supplied to MilkTestNZ (MTNZ). Industry will also collect data to assist in identification of commencement of lactation.

3. For herds that have been supplying continuously (since prior to the 1st July), MTNZ will collate a list of all suppliers as of 1st July, North Island suppliers will be scheduled for 1st sample collection on the 15th August, South Island suppliers will have 1st sample collection on the 30th August.

4. All BTM samples will be tested by PCR; ELISA testing will occur on samples from week 4, 8 and 12.

5. Where a farm tests positive by ELISA:
   a. The ELISA positive milk samples will be forwarded to AHL for repeat ELISA testing, *M. bovis* PCR testing and testing for other Mycoplasma (if *M. bovis* PCR is negative).
   b. The farm will switch to BTM sample collection every other day for a total of 20 PCR samples (40 days).
   c. Despite a positive ELISA result, ELISA tests will continue to be tested as per the schedule in point (1). Additionally:
      i. If the ELISA positive result occurred on BTM sample week 8, the sample at week 12 will be collected and tested by ELISA as planned and a further BTM sample will be collected 4 weeks later (week 16) and tested by ELISA.
      ii. If the ELISA positive result occurred on BTM sample week 12 then 2 additional BTM samples will be collected at 4 week intervals and tested by ELISA (week 16 and week 20).
   d. The objective of additional planned ELISA tests is to detect if the positive ELISA result occurs over multiple occasions.

6. Where a positive ELISA result occurs but all PCR results are negative, further surveillance will be scheduled for that farm. This will be BTM samples collected every 2 weeks and tested by PCR for a total of 6 occasions.
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**Note:** where any ELISA denoted by # test positive sampling will transition to PCR every other day for 40 days (20 samples).
Appendix 2: Sampling flow diagram

**Base surveillance**
Commence 4 weeks after planned start of calving
- PCR test every 2 weeks for 6 tests
- ELISA test every 4 weeks for 3 tests

- **Any PCR positive?**
  - yes
    - Sample from week 4
    - Take remaining base surveillance ELISA samples
  - no
    - Continue base surveillance as planned

- **Any ELISA positive?**
  - yes
    - Sample from week 8
    - Sample from week 12
    - Take further ELISA sample week 12 and week 16
    - Take further ELISA sample week 16 and week 20
  - no
    - Continue base surveillance as planned

- **Status of supplier:**
  - Sampling complete

- **Collect Samples daily for PCR (7 days total)**

- **Continue with protocol as per +ve PCR**

- **Commence PCR sampling every second day for 20 occasions (40 days)**

- **Any positive PCR?**
  - yes
    - Sample sent to MPI AHL for confirmation
  - no
    - Continue base surveillance as planned

- **Samples from week 4**
  - Further ELISA testing depends on which sample tests positive
  - **Any Positive PCR tests already detected?**
    - yes
    - Sample from week 4
    - Sample from week 8
    - Sample from week 12
    - Take further ELISA sample week 12 and week 16
    - Take further ELISA sample week 16 and week 20
    - **Status of supplier:**
      - Sampling complete
    - no
      - Continue with protocol as per +ve PCR

- **Status of supplier:**
  - Enters response
  - **Response**
    - yes
      - Continue base surveillance as planned
    - no
      - **Confirmed?**
        - yes
          - **Status of supplier:**
            - Sampling complete
        - no
          - **Base surveillance**
            - Commence 4 weeks after planned start of calving
            - PCR test every 2 weeks for 6 tests
            - ELISA test every 4 weeks for 3 tests
          - **Any PCR positive?**
            - yes
              - Sample from week 4
              - Take remaining base surveillance ELISA samples
            - no
              - Continue base surveillance as planned
          - **Any ELISA positive?**
            - yes
              - Sample from week 8
              - Sample from week 12
              - Take further ELISA sample week 12 and week 16
              - Take further ELISA sample week 16 and week 20
            - no
              - Continue base surveillance as planned

- **Confirmed?**
  - yes
    - **Status of supplier:**
      - Sampling complete
  - no
    - **Collect Samples daily for PCR (7 days total)**
    - **Continue with protocol as per +ve PCR**
    - **Commence PCR sampling every second day for 20 occasions (40 days)**
    - **Any positive PCR?**
      - yes
        - Sample sent to MPI AHL for confirmation
      - no
        - Continue base surveillance as planned
    - **Status of supplier:**
      - Sampling complete