Self-declaration by Croatia of freedom from bluetongue

Declaration sent to the OIE on 12th of February 2020 by Dr Tatjana Karačić, Assistant Minister, Ministry of Agriculture, Republic of Croatia, OIE Delegate for Croatia.

1. Historical overview and current situation of Bluetongue in Croatia

First documented occurrence of bluetongue disease (BTD) in Croatia was in November 2001 in Dubrovnik-Neretva county, municipality Konavle, the southernmost part of Croatia. Following those outbreaks, serological surveillance started in 2002. Sero-surveillance data until 2005 have demonstrated that virus was circulating among cattle in Dubrovnik-Neretva county (serotype 9 and serotype 16 in the municipalities Metković, Ploče, around Dubrovnik and Konavle). Virus activity during 2006 was limited, and there was no evidence of BTDV circulation from 2007 till October 2014.

In October 2014 first outbreaks of serotype 4 were detected in Dubrovnik-Neretva county.

Altogether 62 outbreaks of bluetongue serotype 4 were confirmed during 2014. The disease was detected in sheep, goats and bovine animals, mostly in small extensive herds. Clinical signs were detected only in sheep and goats. In addition to serotype 4 outbreaks during 2014, one outbreak of Serotype 1 was detected in Šibenik – Knin county. One animal in the flock of 110 animals revealed positive result on serotype 1.

During 2015, 8 limited outbreaks of serotype 1 were detected in Dubrovnik – Neretva county, on island Lastovo (Figure 1 – purple dot). No clinical signs as well as any further spread of that serotype was detected. Last outbreak of BTD in Croatia was confirmed on January 16, 2017.
**Figure 1.** First outbreaks in 2014 of BTD serotype 4 (red dots) and serotype 1 (purple dots)

**Figure 2.** 2016 outbreaks of BTD serotype 4
In total 142 outbreaks were detected from October 2014 to January 2017 involving sheep, 77 goats and 96 bovine animals.

2. Control and eradication measures


- single restriction zone in the whole country
- obligatory vaccination of whole ruminant population
- intensification of active surveillance in the whole country
- protection of animals against vector attacks, where possible and feasible
- outbreak holdings were placed under official control with appropriate
  - movement restrictions
  - epidemiological investigation
  - surveillance
- notification of outbreaks via ADNS and OIE WAHIS

2.1 Vaccination

To control the disease, avoid major epidemic and further spread of the virus, obligatory vaccination campaign on the whole ruminant population started in January 2015. Campaign was financed from the State Budget and co-financed from the European Commission. Mass vaccination that started in 2015 was implemented for 5 consecutive years, until 2019.

Vaccine used: inactivated vaccine against BTD serotype 4.

Only serotype 4 vaccine was used since there was no evidence of circulation or any further spread of serotype 1. After findings of few positive herds in 2015 on island Lastovo, further surveillance did not
detect any serotype 1 infection. All samples positive on PCR that can detect all serotypes from 1-27 are tested additionally with two tests - PCR capable to detect only serotype 4 and only serotype 1, in parallel (see details of testing in point 3).

Bovines, sheep and goats were vaccinated according to manufacturer’s instructions (primo vaccination with booster dose). Vaccination was carried out in the beginning of the year before vector season (January – April). Vaccination was performed by authorised veterinarians. Data on each vaccinated animal are recorded in the National Database.

Vaccination campaign was very successful with very high vaccine coverage.

**Table 1. Details of vaccination campaign 2015 - 2019 – bovine animals**

<table>
<thead>
<tr>
<th>Year</th>
<th>Bovine herds</th>
<th>Bovine herds vaccinated</th>
<th>%</th>
<th>Bovine animals</th>
<th>Bovine animals vaccinated</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>2019</td>
<td>21 178</td>
<td>20 460</td>
<td>97</td>
<td>419 670</td>
<td>401 901</td>
<td>96</td>
</tr>
<tr>
<td>2018</td>
<td>23 220</td>
<td>23 186</td>
<td>100</td>
<td>412 876</td>
<td>408 526</td>
<td>99</td>
</tr>
<tr>
<td>2017</td>
<td>27 236</td>
<td>25 765</td>
<td>95</td>
<td>424 971</td>
<td>402 776</td>
<td>95</td>
</tr>
<tr>
<td>2016</td>
<td>27 904</td>
<td>28 161</td>
<td>101*</td>
<td>418 248</td>
<td>410 465</td>
<td>98</td>
</tr>
<tr>
<td>2015</td>
<td>32 753</td>
<td>30 343</td>
<td>93</td>
<td>431 696</td>
<td>401 593</td>
<td>93</td>
</tr>
</tbody>
</table>

*Data on total number of herds is calculated on December 31st. Number of vaccinated herds is calculated for the whole year and for that reason proportion of vaccinated animals in 2016 is above 100%.

**Table 2. Details of vaccination campaign 2015 - 2019 – small ruminants**

<table>
<thead>
<tr>
<th>Year</th>
<th>Sheep and Goat flocks</th>
<th>Sheep and goats flocks vaccinated</th>
<th>%</th>
<th>Sheep and goats</th>
<th>Sheep and goats vaccinated</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>2019</td>
<td>21 933</td>
<td>16 283</td>
<td>74</td>
<td>767 679</td>
<td>541 388</td>
<td>70</td>
</tr>
<tr>
<td>2018</td>
<td>22 440</td>
<td>19 524</td>
<td>87</td>
<td>743 921</td>
<td>600 586</td>
<td>81</td>
</tr>
<tr>
<td>2017</td>
<td>23 062</td>
<td>20 137</td>
<td>87</td>
<td>727 577</td>
<td>613 080</td>
<td>84</td>
</tr>
<tr>
<td>2016</td>
<td>22 806</td>
<td>19 562</td>
<td>86</td>
<td>707 614</td>
<td>582 157</td>
<td>82</td>
</tr>
<tr>
<td>2015</td>
<td>21 494</td>
<td>21 083</td>
<td>98</td>
<td>705 190</td>
<td>583 870</td>
<td>83</td>
</tr>
</tbody>
</table>

3. Surveillance

The whole susceptible population of animals in Croatia is covered with active and passive surveillance for BTD. Programme is aligned with Articles 8.3.14. – 8.3.17. of the OIE Terrestrial Animal Health Code (Terrestrial Code) and Commission Regulation (EC) No. 1266/2007. The main objectives of the program are to monitor the presence of the disease, to early detect incursion of any other serotype beside BTV S4 and to prove disease absence. In addition to serological surveillance, entomological surveillance was also conducted.

All tests are performed in National Reference Laboratory (NRL) for BTD, which is part of Veterinary Institute Split. Veterinary Institute Split is one of 4 regional branches of Croatian Veterinary Institute Zagreb. NRL performs following tests: VNT, ELISA and PCR tests. PCR tests are as follows: Real time
PCR BTV that can detect all serotypes 1-27, Real time PCR BTV that detects specifically serotype 1 and Real time PCR BTV that detects specifically serotype 4. All samples intended for PCR testing (originating from ELISA positive animals) are first tested with Real time PCR which detects all serotypes from 1-27. In case of positive result, additional PCR for serotypes 1 and 4 are performed in parallel. If results of PCR for serotype 4 and 1 would be negative, samples would be sent for further testing to EURL for BTD.

Sampling is performed by authorised veterinary organisations. Authorised veterinary organisations are private veterinary organisations, under contract with Ministry of Agriculture, Veterinary and Food Safety Directorate and supervision of veterinary inspectors from State Inspectorate.

### 3.1 Active surveillance

Active surveillance for BTD was conducted for many years, following first BTD outbreaks in 2001. From 2014 surveillance was intensified. Testing is performed using ELISA test. In case of ELISA positive animals, further testing with PCR was conducted. From 2015 and onwards testing was adjusted to the vaccination campaign and performed on non-vaccinated bovine animals (sentinels). In case of insufficient number of bovine animals, goats or sheep were selected. Detailed surveillance results are presented in Table 3. and in maps presenting geographical distribution of samples in 2018 and 2019.

In 2018, from total number of 2481 ELISA samples, 796 were originating from bovine animals, 1655 from sheep and 30 from goats. In 2019, 2326 ELISA samples were originating from bovine animals and 221 from sheep. Sample size was planned in order to detect disease if it was present in 5 % prevalence, with CI 95 %. Sampling was organised in administrative counties, each representing one or more geographical units (size of geographical unit 45*45 km2). Very high vaccination coverage and small animal density in certain counties created some difficulties in achieving planned number of sentinel animals.

First goal of testing was to monitor the disease presence in the country. For that reason, animals are tested during the time of the year when infection is most likely to be detected (testing two times, in August/September and again on November/December). For this purpose, animals from the whole country were selected.

Second goal of surveillance was aimed to early detect incursion of any other serotype other than BTV4, in areas which are at risk of such introduction. For this purpose, the whole coastal area of Croatia is considered to be at risk for BTV1 introduction from Italy. Sampling was also performed during the time of the year when infection is most likely to be detected (testing from August/September to December).

### Table 3. Details of active surveillance 2015 - 2019

<table>
<thead>
<tr>
<th>Year</th>
<th>Herds tested</th>
<th>Animals tested</th>
<th>ELISA tests</th>
<th>ELISA positive samples</th>
<th>PCR tests</th>
<th>PCR positive samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>2019</td>
<td>817</td>
<td>1367</td>
<td>2547</td>
<td>229</td>
<td>186</td>
<td>0</td>
</tr>
<tr>
<td>2018</td>
<td>616</td>
<td>641</td>
<td>2481</td>
<td>313</td>
<td>246</td>
<td>0</td>
</tr>
<tr>
<td>2017</td>
<td>712</td>
<td>1020</td>
<td>2549</td>
<td>397</td>
<td>295</td>
<td>1</td>
</tr>
<tr>
<td>2016</td>
<td>1233</td>
<td>2001</td>
<td>3872</td>
<td>618</td>
<td>577</td>
<td>65</td>
</tr>
<tr>
<td>2015</td>
<td>563</td>
<td>1982</td>
<td>5361</td>
<td>1249</td>
<td>643</td>
<td>99</td>
</tr>
</tbody>
</table>
Last PCR positive sample was confirmed on 16 January 2017. Total number of samples / positive ELISA or PCR samples in the period 2015 – 2019 is presented in Annex 1. Figures 4. and 5. Geographical distribution of samples for 2018 and 2019 is presented in Annex 2.

3.2 Passive surveillance

BTD is notifiable disease in the whole country. The Ordinance on the notification of animal diseases (Official Gazette 135/14) sets out the obligation to notify the occurrence and suspicion of BTD and lays down the procedures to be followed by the keeper of the animal, the authorised veterinarian, the veterinary inspector and official laboratory. The keeper of the animal must immediately and without delay notify the authorised veterinary organisation on suspicion of the disease (clinical signs). In case animal shows clinical signs of BTD, blood samples for serological and virological testing are taken by an authorised veterinarian and investigated in the National Reference Laboratory for BTD (Croatian Veterinary Institute – regional branch in Split).

During 2018, there were 3 flocks in which animals had clinical signs that could suggest possible infection with BTD virus (high fever, tongue oedema, nasal discharge). All animals were PCR negative. There was no reporting of clinical signs that could suggest possible infection with BTD in 2019.

3.3 Entomological surveillance

First entomological surveillance was conducted from 2009 to 2011. Sampling for vector presence was performed by authorised veterinary organisations on randomly selected locations. It was concluded that adult insects from family Culicoides are present during the whole year, but also, that they are significantly reduced during winter months. Highest vector activity is from June to September. Entomological surveillance started again from 2015 with similar results. It is conducted on predefined locations in all 21 counties. Samples were collected once a month from February to December. Due to climate conditions and detection of vectors throughout the year (with reduction during December and January) vector free period was not declared.

4. Measures for maintenance of status

4.1 Early detection system

4.1.1 Surveillance


4.1.2 Notification

BTD is notifiable disease in Croatia according to the Ordinance on the notification of animal diseases (Official Gazette 135/14) which sets out in detail the obligation to notify the occurrence and suspicion of BTD. Ordinance lays down the procedures to be followed by the keeper of the animal, authorised veterinarian, veterinary inspector and official laboratory. The keeper of the animal must immediately and without any necessary delay notify suspicion of the disease (clinical signs) to veterinarian. A veterinarian who suspects the disease must notify the Veterinary and Food Safety Directorate and the veterinary inspector by telephone or by electronic means, without delay and not later than within 24 hours.
4.2 Movement rules

Transport of susceptible animals and their germinal products from restricted areas to other member states has to comply with Annex III of Commission Regulation (EC) No. 1266/2007. Safe movement of animals stipulated in Regulation 1266/2007 is based on movements of immunised animals, either due to vaccination or natural immunity. This movement is considered safe irrespective of the virus circulation at the place of origin or the vectors activity at the place of destination. In addition to immunised animals, safe movement of animals is also based on protecting animals from vector attacks, followed by negative PCR result before movement.

The requirements for the import from non-EU countries are laid down in Commission Regulation (EU) No 206/2010 laying down lists of third countries, territories or parts thereof authorised for the introduction into the European Union of certain animals and fresh meat and the veterinary certification requirements. Regulation 206/2010 lays down the lists based on the principles contained in Council Directive 2004/68/EC. The most important aspects are the health status of livestock, legislation of the non EU country, rules on the prevention and control of animal diseases, organization, structure, competence and power of the veterinary services, membership of the World Organization for Animal Health (OIE) and the regularity and rapidity of information on infectious animal diseases to the Commission and the OIE.

5. Conclusions

Based on the information provided in this report and in accordance with the provisions of Article 8.3.3. of the OIE Terrestrial Code, this self-declaration provides documented evidence that:

- Last outbreak of BTD in Croatia was confirmed on 16 January 2017
- Vaccination campaign notably reduced viral circulation in the country and further spread of the virus
- The results of active and passive surveillance demonstrate that there is no evidence of infection with BTD virus in Croatia for more than two years.
- Surveillance and vaccination strategies are supported by entomological surveillance.

Consequently, the OIE Delegate of Croatia declares that the country fulfils the requirements for a freedom from bluetongue disease as of 1 January 2020, in compliance with Chapter 1.6. and Article 8.3.3. of the OIE Terrestrial Code (2019) and consistent with the information provided in WAHIS.

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Annex 1. Testing results for the period 2015 – 2019

Figure 4. ELISA tests results 2015 – 2019

Figure 5. PCR tests results 2015 – 2019

Map 1 – geographical distribution of samples in 2018

Map 2 – geographical distribution of samples 2019