Bluetongue in Italy: Part II

A. Giovannini(1), P. Calistri(1), D. Nannini(1), C. Paladini(1), U. Santucci(2), C. Patta(3) & V. Caporale(1)

(1) Istituto Zooprofilattico Sperimentale dell’Abruzzo e del Molise ‘G. Caporale’, Via Campo Boario, 64100 Teramo, Italy
(2) Ministero della Salute, Direzione Generale della Sanità Pubblica Veterinaria, Alimenti e Nutrizione, Piazzale Marconi, 00144 Rome, Italy
(3) Istituto Zooprofilattico Sperimentale della Sardegna ‘G. Pegreffi’, Via Duca degli Abruzzi N° 8, 07100 Sassari, Italy

Summary

In summer 2000, bluetongue (BT) infection was reported in Italy and caused a widespread epidemic involving a total of ten southern and central regions and is still in progress after three years. From the date of the first case (18 August 2000) to 14 May 2001, when the lowpoint in the first epidemic curve was reached, a total of 310,234 animals in 6,869 flocks of three regions had been involved. From 15 May 2001 to 14 April 2002, when a second epidemic wave swept through central and southern Italy, a total of 323,635 animals in 6,807 flocks in seven regions were involved. During 2000 and 2001 virtually no susceptible ruminants were vaccinated. On 11 May 2001, the Italian Ministry of Health ordered the vaccination of all susceptible domestic ruminant species (i.e. sheep, goats, cattle and water buffalo) in the infected and surrounding areas. The vaccination strategy stemmed from a risk assessment that demonstrated the possibility of such a strategy preventing most of the direct economic losses and decreasing the level of virus circulation. Vaccination of the target populations commenced in January 2002. In July 2002, when the new epidemic peak was reached, the percentage of vaccinated populations varied between the regions with direct consequences on the spread of BT. The relationship between vaccination coverage of the target populations and animal losses due to disease and virus circulation, and as detected by the sentinel surveillance system, was analysed. The effectiveness of the vaccination campaign in limiting virus circulation and consequently indirect losses due to animal movement restrictions was analysed and evaluated. At the end of 2002, a second risk assessment led to the authorisation of the movement of vaccinated animals from infected areas (where at least 80% of the susceptible population was vaccinated) directly to slaughter in unvaccinated areas free from infection. This risk assessment also generated new criteria to define zones where animal movement restrictions should be applied. Following the second vaccination campaign (January to May 2003), a third risk assessment was performed and the results from vaccination trials performed in controlled and in field conditions studied. These studies indicated that procedures to move vaccinated breeding animals from zones where infection exists to unvaccinated infection free zones could be contemplated.

Keywords


Introduction

In August 2000, bluetongue (BT) infection, due to serotype 2, was reported for the first time on mainland Italy, in the Balearic Islands (Spain), and in Corsica (France). By the autumn of 2000, a second serotype, BT virus (BTV) serotype 9, was observed in southern Italy. This incursion of BTV into the central Mediterranean region resulted in the largest epidemic of BT ever to affect Europe.

Since 2000, BT has spread to a total of ten southern and central regions of Italy. In the first epidemic commencing on 18 August 2000 and ending on 14 May 2001, a total of 262,759 animals in 6,869 flocks in three regions were affected. From 15 May 2001 to 14 April 2002, a second epidemic wave swept through central and southern Italy, affecting a total of 250,662 animals in 6,807 flocks in seven regions (1).
Following the first epidemic, a risk assessment indicated that it would be possible to prevent most direct economic losses by vaccinating sheep. Virus circulation could also be reduced if all domestic ruminant populations susceptible to BTV infection (sheep, goats and cattle) were vaccinated (4). Consequently, the Italian Ministry of Health ordered the vaccination of all domestic ruminants susceptible to BT infection (sheep, goats, cattle, buffaloes) in infected and in at-risk areas on 11 May 2001 (7). Despite the Order of the Ministry, virtually no ruminants were vaccinated during 2001. Vaccination of susceptible populations commenced in January 2002 and when the new seasonal epidemic started (July 2002), the level of vaccination in susceptible populations varied greatly in the different regions of Italy. Differences in vaccination coverage of susceptible domestic populations resulted in different levels of loss due to the disease and to different intensities of virus circulation and spread.

The objectives of the study is to describe the third and fourth seasonal epidemics in Italy, after the implementation of a vaccination strategy, and to analyse the effectiveness of vaccination in reducing both the direct losses due to the disease and the indirect losses due to restrictions of movements of animals from infected areas.

**Methods**

**Source of data**

The data on BT outbreaks, serological surveillance of sentinel animals, entomological surveillance, ad hoc surveys and vaccination activities recorded in the Bluetongue National Information System (6) are analysed here.

**Outbreaks**

When a case of BT was suspected to occur on a farm(s), all susceptible animals were examined for clinical signs by official veterinarians and blood samples collected and sent to the laboratory for confirmation of infection. Morbidity and mortality was monitored and recorded during weekly visits to affected farms. When BT was confirmed, official veterinarians extended visits to all ovine flocks within a radius of 20 km of the confirmed outbreak (when clinical disease was observed) or to all ovine flocks within a radius of 4 km (if there was subclinical infection).

**Serological surveillance of sentinel animals**

A nationwide sentinel network had already been implemented in January 2002 when the vaccination campaign commenced. Italy had been divided into a grid of square units of either 400 km$^2$ or 1 600 km$^2$ depending on the occurrence, or the risk of occurrence, of infection. To detect at least 5% of positive animals with a 95% confidence level in each 400 km$^2$ unit, a sample of 58 bovine animals was selected from 5 to 8 farms. To detect at least 2% of positive animals with a 95% confidence level in each unit of 1 600 km$^2$, a sample of 148 bovine animals was selected from 8 to 12 farms. If cattle were not present in the area, sheep were selected as sentinel animals. Blood samples from sentinels were collected regularly with a variable frequency dependent upon the season and infection occurrence in the area.

**Entomological surveillance**

Nationwide entomological surveillance was also implemented before vaccination started. Blacklight traps were positioned in fixed locations in each province and operated weekly to monitor Culicoides spp. population dynamics. Blacklight traps were also operated on a temporary basis in suspected or confirmed cases of virus circulation to identify the Culicoides spp. present.

**Vaccination**

To monitor the progress of the vaccination campaign, vaccine serotype(s), vaccine batch numbers, farm codes, total numbers of animals on the farm, numbers of eligible animals and numbers of vaccinated animals by species, and vaccination date were recorded by the Local Health Unit. Data were then sent to the Bluetongue National Information System where it was sorted by province and circulated on the internet.

Sera from 35 randomly selected vaccinated animals per each grid cell were tested for antibody presence to monitor vaccination coverage. Antibody coverage in vaccinated populations was evaluated by serological examination of the sample collected.

Possible undesirable vaccine side-effects (deaths, abortions and stillbirths), were monitored by:

a) sampling animals in flocks where problems arose to assess the presence of the vaccine virus in dead animals and/or foetuses

b) collecting information concerning the type and incidence of the loss observed, vaccine used, dates of vaccination, etc.

Samples were submitted to the laboratory to perform differential diagnostic tests and for the identification of the BTV serotype involved (vaccine or wild-type virus).
Analysis of data

Efficacy of vaccination in reducing direct losses due to the disease in the regions affected was evaluated by comparing the percentage of the population vaccinated at the beginning of the new epidemic (July 2003) with the geographic distribution of the infection and the number of outbreaks during the 2002 epidemic. The correlation between the percentage of the population vaccinated by the end of July 2002 and the number of outbreaks in the 2002-2003 epidemic was evaluated using Spearman’s correlation coefficient (11).

The effectiveness of vaccination in reducing indirect losses due to movement restrictions was evaluated in Sardinia. The number of animals exported from Sardinia to free areas in continental Italy in 2001 (before the commencement of the vaccination campaign) was compared to the number in 2002 (after vaccination had been completed).

The decrease in the proportion of vaccinated animals in the population due to replacements was also estimated, assuming a regular yearly 20% replacement rate.

Results and discussion

Evolution of the epidemic

The third BT epidemic started in Italy on 15 April 2002 and ended on 14 April 2003. During this outbreak, BTV-2 and BTV-9 infection spread to the Province of Avellino (Campania) in July and to the Provinces of Benevento and Caserta (Campania), Foggia and Bari (Apulia), l’Aquila (Abruzzi) and Isernia (Molise) in September. The only spread of BTV-2 infection to a new zone was observed in Massa (Tuscany) in September. The total number of outbreaks detected in the third epidemic was 432 in eight regions (Table I). The geographical distribution of the infection is presented in Figure 1.

Moreover, during the third epidemic, two new serotypes were detected in southern Italy, namely: BTV-4 and BTV-16. Both serotypes had been reported previously in the eastern Mediterranean and in Greece. The source of the spread of these serotypes to Italy is still unknown, but illegal trade in animals is suspected.

<table>
<thead>
<tr>
<th>Region</th>
<th>Number of outbreaks</th>
<th>Total number of animals in infected flocks</th>
<th>Number of diseased animals</th>
<th>Number of dead animals</th>
<th>Number of slaughtered animals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basilicata</td>
<td>59</td>
<td>5 145</td>
<td>223</td>
<td>96</td>
<td>–</td>
</tr>
<tr>
<td>Calabria</td>
<td>15</td>
<td>797</td>
<td>87</td>
<td>–</td>
<td>87</td>
</tr>
<tr>
<td>Campania</td>
<td>251</td>
<td>20 918</td>
<td>1 951</td>
<td>1 495</td>
<td>213</td>
</tr>
<tr>
<td>Lazio</td>
<td>14</td>
<td>1 702</td>
<td>44</td>
<td>37</td>
<td>–</td>
</tr>
<tr>
<td>Molise</td>
<td>13</td>
<td>2 781</td>
<td>5</td>
<td>8</td>
<td>–</td>
</tr>
<tr>
<td>Puglia</td>
<td>17</td>
<td>2 484</td>
<td>284</td>
<td>245</td>
<td>1</td>
</tr>
<tr>
<td>Sardinia</td>
<td>10</td>
<td>2 120</td>
<td>28</td>
<td>5</td>
<td>4</td>
</tr>
<tr>
<td>Sicilia</td>
<td>53</td>
<td>12 304</td>
<td>1 076</td>
<td>1 092</td>
<td>–</td>
</tr>
<tr>
<td>Total</td>
<td>432</td>
<td>48 251</td>
<td>3 698</td>
<td>2 978</td>
<td>305</td>
</tr>
</tbody>
</table>
During the summer of 2003, the fourth epidemic commenced and clinical disease was observed in Sardinia alone in August involving BTV-4, a serotype that had not been included in the vaccination programme. By 7 October 2003, 850 new outbreaks had occurred in southern and western Sardinia, following a pattern that closely resembled that recorded during the summer of 2000 (Table II). No disease was recorded in central and southern Italy, but seroconversions in sentinel animals did occur (Fig. 2).

Table II

<table>
<thead>
<tr>
<th>Region</th>
<th>Number of outbreaks</th>
<th>Animals in infected flocks (total)</th>
<th>Number of diseased animals</th>
<th>Number of dead animals</th>
<th>Number of slaughtered animals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sardinia</td>
<td>850</td>
<td>241 648</td>
<td>648</td>
<td>1 585</td>
<td>9</td>
</tr>
</tbody>
</table>

Vaccination

Vaccination in Italy was performed using two different vaccines, depending on the BTV serotypes observed in the various zones, namely: a monovalent BTV-2 vaccine was used in Sardinia, Tuscany and Latium, and a bivalent vaccine (BTV-2 and BTV-9) was used in the southern regions. In 2002, zones in which vaccination had been practised were modified according to the spread of infection (Fig. 3).

Vaccination in infected zones and in areas at risk commenced on a limited scale in the late autumn of 2001. In most regions and provinces involved the vaccination programme commenced in January 2002 (Fig. 4). When the new epidemic began, in July 2002, 57% of the eligible animals in Italy had already been vaccinated (Fig. 4) but vaccination coverage in the various regions varied greatly (Figs 5, 6, 7, 8, 9, 10 and 11). Sardinia (Fig. 5) and Tuscany (Fig. 6) were able to vaccinate approximately 90% (97% in Sardinia and 87% in Tuscany) before the commencement of the new epidemic. In Basilicata, on the other hand (Fig. 7) only 2% of the population was vaccinated before the new epidemic started in July 2002 and only by the end of December 2002 had 84% of the eligible population been vaccinated. In the other regions (Sicily, Latium, Calabria and Campania) (Figures 8, 9, 10 and 11), less than two-thirds of the populations were vaccinated.

Direct losses: number of outbreaks and diseased animals

The different levels of vaccination had clear consequences on disease occurrence. In the two regions in which approximately 90% of the ruminant population was vaccinated, clinical disease either disappeared (Tuscany: 158 outbreaks and 693 diseased animals in the 2001-2002 epidemic, 0 outbreaks in 2002-2003) (Fig. 6) or was reduced by a factor of 1/100 (Sardinia: 6 090 outbreaks and 239 178 diseased animals in the 2001-2002 epidemic, 10 outbreaks and 28 diseased animals in 2002) (Fig. 5). In these regions, the spread of infection was also significantly reduced by the vaccination campaign (Fig. 12). A clear demonstration of the effectiveness of vaccination was observed in Sardinia where, in August 2003, a new epidemic due to BTV-4 started, causing 850 outbreaks in six weeks; this would seem to demonstrate that the decrease in BTV-2 circulation was due to vaccine-induced resistance and not to the disappearance of conditions favouring the spread of the virus.
outbreaks recorded in the 2002-2003 epidemic in the five regions of central and southern Italy was significantly correlated to the level of vaccination achieved by each region at the end of July 2002 (Spearman’s $\rho = -0.9150, p<0.0001$).

**Indirect losses: the export of cattle from Sardinia to continental Italy**

The vaccination of ruminant populations was conducive to a progressive reduction of virus circulation and consequently of the zones in which movement restrictions were applied. Sardinia has been taken as an example to evaluate the effect of vaccination on animal trade for the following reasons:

a) before the BT epidemic, cattle were traded extensively between Sardinia and continental Italy, especially northern Italy

b) after the appearance of the disease in Sardinia, the export of cattle from the island to disease-free areas in continental Italy came to a complete standstill

c) the progressive relaxation of movement restrictions paved the way for the resumption of exports to disease-free areas in northern Italy.

---

**Figure 3**

Changes in the distribution and numbers of provinces (in brackets) vaccinated in Italy, 2002
Figure 4
Monthly curve of animal populations vaccinated and outbreaks of bluetongue in Italy, August 2000-October 2003

Figure 5
Monthly curve of animal populations vaccinated and outbreaks of bluetongue in Sardinia, August 2000-October 2003

Figure 6
Monthly curve of animal populations vaccinated and outbreaks of bluetongue in Tuscany, August 2000-October 2003

Figure 7
Monthly curve of animal populations vaccinated and outbreaks of bluetongue in Basilicata, August 2000-October 2003

Figure 8
Monthly curve of animal populations vaccinated and outbreaks of bluetongue in Sicily, August 2000-October 2003

Figure 9
Monthly curve of animal populations vaccinated and outbreaks of bluetongue in Lazio, August 2000-October 2003
The total number of bovine animals sent from Sardinia to continental Italy in 2001 was 1 019 (Fig. 13). Of these, 92% were moved during the last two months of the year when the effects of vaccination were visible and when extensive areas of northern Italy were free from vectors and could consequently receive animals from the surveillance zones without the risk of losing their free status (2, 9). Towards the end of 2002, a new risk assessment (3, 5) led to the authorisation of movements (for direct slaughter) of vaccinated animals from infected areas where at least 80% of the susceptible population has been vaccinated; the risk assessment also led to a new approach to define those areas under movement restrictions. From January to June 2002, a total of 3 097 cattle were exported from Sardinia to continental Italy, compared to eight animals in the same period of the previous year. The arrival and spread of BTV-4 to most of the island in August 2003 once again brought the exports from Sardinia to a new halt.
serotypes that are administered concurrently. Based on the results of these trials, and upon the level of efficacy achieved in the second vaccination programme (January-May 2003), a further risk assessment will be performed. Its primary aim will be to establish whether it is feasible to trade in vaccinated animals originating from areas in which BTV is still actively circulating. This finding could contribute to further revision of current trade regulations.

References


