

# Analysis of climatic factors involved in the BTV-1 incursion in Central Italy in 2014

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## Keywords

Anomalies,  
Bluetongue virus  
serotype 1,  
Climate,  
*Culicoides*,  
Rainfall,  
Temperature.

## Summary

In 2012, six years after the previous epidemic, Bluetongue virus serotype 1 (BTV-1) re-emerged in Sardinia causing a limited number of outbreaks. Due to impossibility of implementing a vaccination campaign, the BTV-1 then spread all over the island in 2013 with about 7,000 outbreaks and, in September 2013, the virus reached Central Italy, with a limited number of outbreaks located along the Tyrrhenian coast. The surveillance system in place in Italy detected viral circulation during the following winter, when a few seroconversions were notified. Starting from mid July 2014, a huge number of outbreaks were reported and the disease spread toward inland territories, affecting Umbria, Abruzzo and Marche. In 2014, BTV-1 affected areas where *Culicoides* species belonging to the *Obsoletus* and *Pulicaris* complexes were identified as main vectors. The analysis of temperature and rainfall in Central Italy revealed a significant warmer winter (2013-2014) and a cooler and rainy summer season (2014). These climatic aspects might have certainly favored the overwintering of the virus in local vector or host populations in the Tyrrhenian coast, and, then, the spread of the virus to the rest of Central Italy. However, the heavy circulation of BTV-1 and the severity of clinical outbreaks recorded leave a number of 'open questions' that are currently under investigations.

## Associazione tra variabili climatiche e incursione di BTV-1 in Italia centrale nel 2014

## Parole chiave

Anomalie,  
Clima,  
*Culicoides*,  
Pioggia,  
Sierotipo 1 del virus  
della Bluetongue,  
Temperatura.

## Riassunto

Nel 2012, a 6 anni dalla precedente epidemia, il sierotipo 1 del virus della Bluetongue (BTV-1) è riapparso nel sud della Sardegna provocando un numero limitato di focolai. L'impossibilità di una tempestiva vaccinazione ha causato il diffondersi del BTV-1 su tutta l'isola nel corso del 2013 provocando circa 7.000 focolai e, a settembre 2013, il virus ha raggiunto l'Italia centrale. L'arrivo a fine stagione, con il calo delle temperature, ha impedito una estesa diffusione del virus, che ha interessato solo alcune aree della costa tirrenica. Durante l'inverno, il sistema di sorveglianza nazionale basato su animali sentinella ha rilevato circolazione virale in alcune aree della regione Lazio. L'estate 2014 ha registrato la più grande epidemia verificatasi fino ad oggi in Italia centrale, con il coinvolgimento di Umbria, Marche e Abruzzo. I principali vettori presenti nell'area e responsabili della diffusione appartengono ad *Obsoletus* e *Pulicaris* complexes. L'analisi di dati climatici di temperatura e pioggia nell'area interessata dai primi focolai ha evidenziato un inverno più mite (2013-2014) rispetto al trend degli ultimi anni e un'estate più fresca e caratterizzata da abbondanti piogge (2014). Questi fattori potrebbero aver favorito l'overwintering del virus nei vettori locali o nelle popolazioni ospite e aver successivamente contribuito all'estesa diffusione durante l'estate 2014. La massiccia circolazione virale e la severità dei focolai clinici lasciano una serie di "domande ancora aperte" per le quali sono in corso più dettagliate ricerche.

## Introduction

Bluetongue virus (BTV) is the etiological agent of Bluetongue (BT), an infectious, non-contagious disease of domestic and wild ruminant, that is transmitted by *Culicoides* biting midges (Diptera: Ceratopogonidae). Before 1998, BT has occurred sporadically in Europe, with a main BTV-10 outbreak on the Iberian peninsula between 1956 and 1960 (Manso-Ribeiro *et al.* 1957) and a minor BTV-4 outbreak on Greek islands in 1979 (Vassalos 1980). Since 1998, a dramatic change in the BT scenario was observed in Europe with several incursions of different strains belonging to different BTV serotypes (Lorusso *et al.* 2013). The circulation of BTV is confined to those periods of the year when the climatic conditions are favorable for adult vector activity and for virus replication in the vectors (Mellor and Leake 2000, Purse *et al.* 2005, Sellers and Mellor 1993).

Due to its peculiar location in the middle of the Mediterranean basin, Italy, since the first introduction of Bluetongue virus in August 2000 (Calistri *et al.* 2004), experienced BTV incursions almost constantly every year. Six different serotypes (BTV-1, BTV-2, BTV-4, BTV-8, BTV-9, BTV-16) spread in different regions between 2000 and 2014 (Lorusso *et al.* 2014, Lorusso *et al.* 2013). Massive vaccination campaigns have prevented the virus from spreading in the Italian territory, however when they were not properly implemented, the virus spread, overwintered both in hosts or vectors (Mayo *et al.* 2014, Takamatsu *et al.* 2003), and re-emerged in the next season.

In 2012, six years after the previous epidemic, BTV-1 re-emerged in Sardinia (Lorusso *et al.* 2013), causing 421 outbreaks in the southern part of the island. It was not possible to implement a vaccination campaign

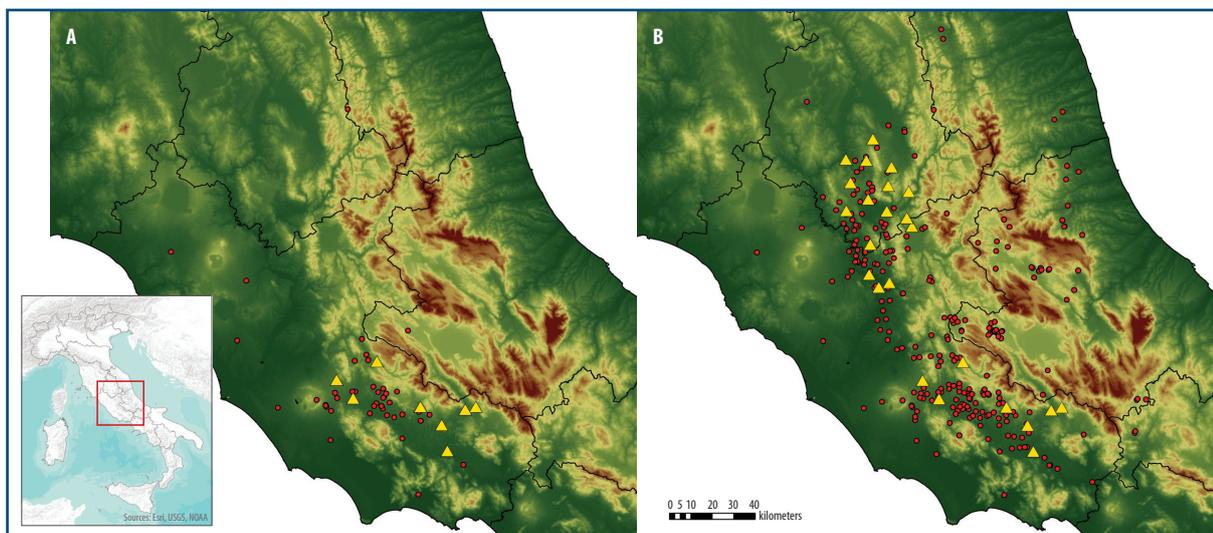
due to the lack of vaccines and in the following year BTV-1 spread all over the island causing about 7,000 outbreak. On the 29<sup>th</sup> of September 2013 it reached Central Italy. About 70 outbreaks occurred, mainly located along the Tyrrhenian coast of Lazio and Tuscany where both species of the *Obsoletus* complex and *Culicoides imicola* are present (Goffredo *et al.* 2004, De Liberato *et al.* 2005). During the winter, the national surveillance system based on sentinel animals kept detecting virus circulation in the area, even if no further spatial distribution of outbreaks was reported. In summer 2014, the disease spread to Umbria, Abruzzo and Marche, affecting areas where the majority of *Culicoides* associated to livestock belonged to the *Obsoletus* complex and, to a lesser extent, to the *Pulicaris* complex (De Liberato *et al.* 2005, Goffredo *et al.* 2015).

Aim of the paper is to investigate and describe the climatic factors possibly associated with the overwintering of the virus and with the exceptional spread of the infection which occurred in summer 2014. Temperature and rainfall collected in the area of first infection (in Terni, Rieti, Roma, Viterbo and Frosinone provinces), were investigated and possible consequences to the potential vector cycle are discussed.

## Materials and methods

### Study area

The evaluation of climatic variables was conducted in a study area located in Lazio and Umbria, the first two regions being largely affected by BTV-1. Figure 1 (A and B) shows the outbreak spatial distribution from the 1<sup>st</sup> of January 2014 to the 1<sup>st</sup> week of



**Figure 1. A.** Bluetongue virus serotype 1 (BTV-1) outbreaks distribution (in red) from the 1<sup>st</sup> of January 2014 to the end of July 2014. **B.** BTV-1 outbreaks distribution (in red) from 1<sup>st</sup> of January to the 1<sup>st</sup> week of September 2014. In yellow the DEWTRA weather stations.

September 2014 and the weather stations used to collect climatic data.

### Surveillance data and climatic data

Data on BTV circulation include both clinical cases and seroconversions in sentinel animals as notified in 2013 and 2014 to the National Information System for Disease Notification (Iannetti *et al.* 2014) of the Ministry of Health<sup>1</sup>. Entomological data are collected in the frame of the Entomological Surveillance National Plan for bluetongue.

Daily minimum and maximum temperature and rainfall from 2007 to 2014 were provided by the DEWETRA platform, a real-time integrated system for hydro-meteorological and wildfire risk forecasting, monitoring and prevention from the National Department for Civil Protection.

### Statistical analysis

To verify the presence of anomalies of temperature and rainfall, climatic daily data were aggregated at weekly level (average for temperatures and cumulated for rainfall). For each week in the period 2007-2014, anomalies were calculated by z-scores using the interquartile range as variability measure (Ministry of Agriculture, Fisheries and Food, Norfolk, UK., 1993):

$$Z_{ij} = \frac{x_{ij} - \bar{x}_i}{0.7413 \cdot IQ_i}$$

Where :

$x_{ij}$  is the climatic value in the  $i^{\text{th}}$  week in the year  $j$ ;

$\bar{x}_i$  is the average value in the  $i^{\text{th}}$  week of all the weather stations in the entire period 2007-2014;

$IQ_i$  is the interquartile range in the  $i^{\text{th}}$  week in the period 2007-2014.

To detect longer periods with values systematically above or below the average, the Rescaled Sum of Scores (RSZ) (Guide to NATA Proficiency Testing 1977) was calculated for each week  $i$  and year  $j$  considering a moving window of five weeks:

$$RSZ_{i,j} = \frac{\sum_{i-4}^i Z_{i,j}}{\sqrt{5}}$$

An anomaly is defined as any value with an absolute z-score  $> 3$  (i.e.  $|z| > 3$ ), thus having less than 1% of chance to consider an outlier as a true member of the series. In the case of z-score between  $\pm 2$  and

$\pm 3$  (i.e.  $2 < |z| < 3$ ), the probability of error in the recognition of anomalies is between 1% and 5%. The interpretation of RSZ follows the same criteria with 5 weeks aggregation period.

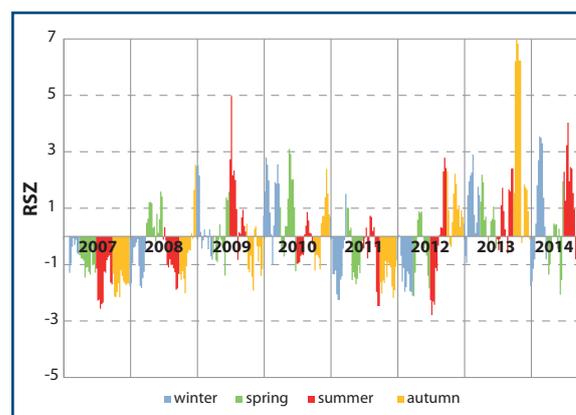
### Results

Figure 1 shows the study area and the BTV-1 circulation from the 1<sup>st</sup> of January 2014 to the end of July 2014 (A) and to the 1<sup>st</sup> week of September 2014 (B). The 25 DEWTRA weather stations used to collect climatic data are showed in yellow.

The analysis of the anomalies in the past 7 years in the study area, revealed a systematic pattern of higher level of rain since summer 2012 with significant high values in autumn 2013 (Figure 2). For temperatures, no systematic pattern was evident, but the analysis revealed a significant warmer winter in 2013-2014 (RSZ for temperature values higher than 2) and cooler 2014 summer (RSZ for temperature values less than -2). In particular, the minimum temperature was systematically higher during the Winter (Figure 3A) and the maximum temperature was systematically lower during the summer (Figure 3B).

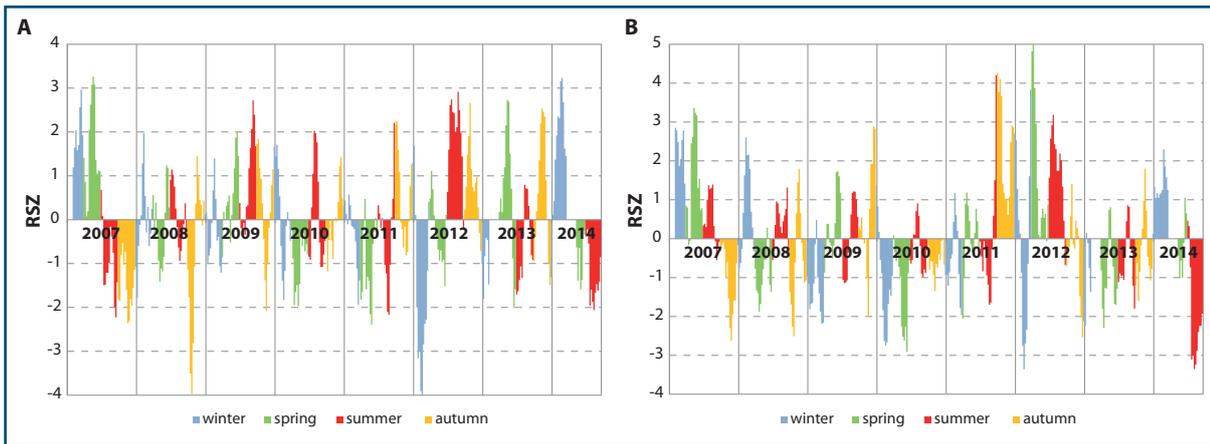
### Discussion

Since 2000, when BT firstly appeared on the Italian territory, several epidemics have reached Central Italy, but the spread remained clustered along the Tyrrhenian coast in Tuscany and Lazio (Calistri *et al.* 2004). During 2013, the BTV-1 spread across the entire Sardinia and in September 2013 (Figure 4) it reached the Tyrrhenian coast of Central Italy. In the following months of 2013, the infection remained clustered along the coast, without further spread to other inland territories, except for a few spots (Figure 5). The limited spread of BTV-1 in 2013 (with only 68 outbreaks) was probably because the virus

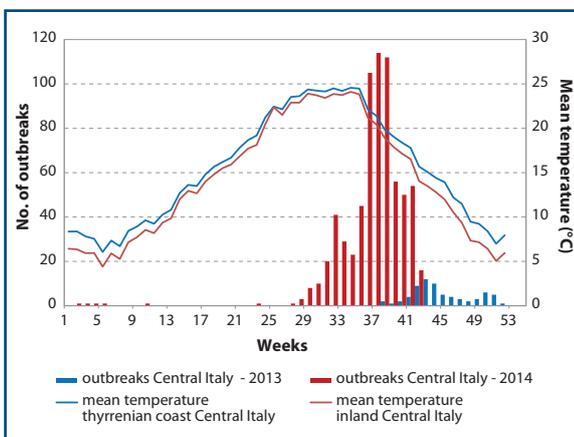


**Figure 2.** Anomalies in cumulated weekly rainfall recorded between 2007-2014 in the epidemic area early affected by Bluetongue virus serotype 1 in 2014 (RSZ = Rescaled Sum of Scores).

<sup>1</sup> <https://www.vetinfo.sanita.it>.



**Figure 3.** Anomalies in weekly temperature [minimum temperature (A), maximum temperature (B)] recorded between 2007-2014 in the study area.



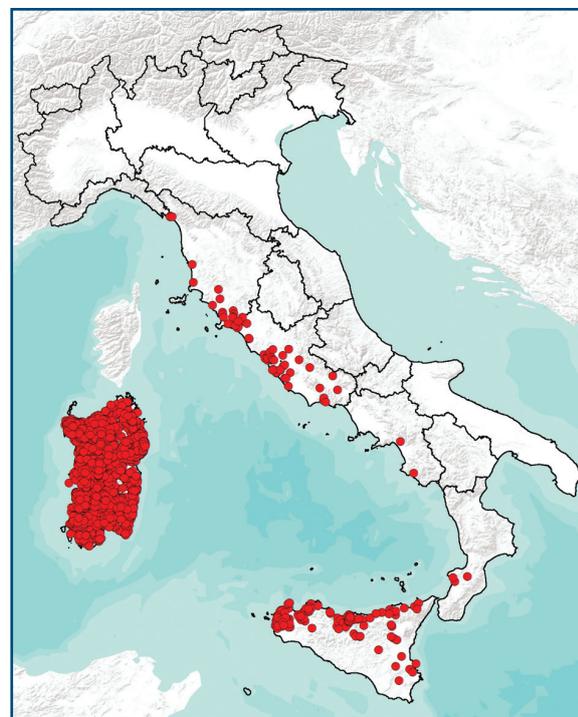
**Figure 4.** Epidemic curves of 2013 and 2014 Bluetongue virus serotype 1 in Central Italy and mean temperature trends (temperatures provided by the DEWETRA platform).

arrived on the area when temperatures were not favorable to the vector capacity (Figure 4).

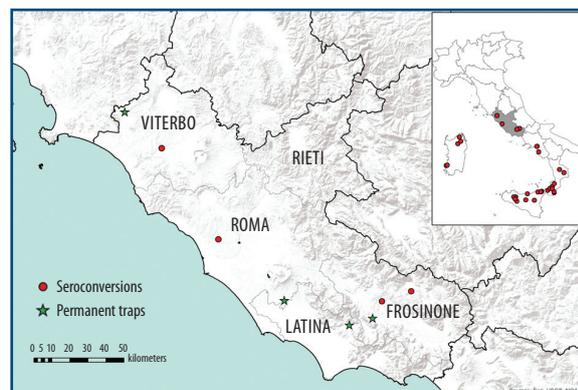
From late December 2013 to March 2014, in four farms, investigated in the frame of the serological national plan, sentinel animals seroconverted to BTV-1, indicating the continuous virus circulation in the area during the winter/spring season (Figure 4 and 6).

The results of the climatic investigation of temperature and rainfall anomalies in the study area (area of possible overwintering), indicated, in autumn 2013, a rainy pattern higher than the average and a 2013-2014 winter season significantly warmer than the trend of the period. This pattern is in line with what observed all across Europe where the winter 2013-2014, has been recorded as the third warmest since 1950<sup>2</sup>.

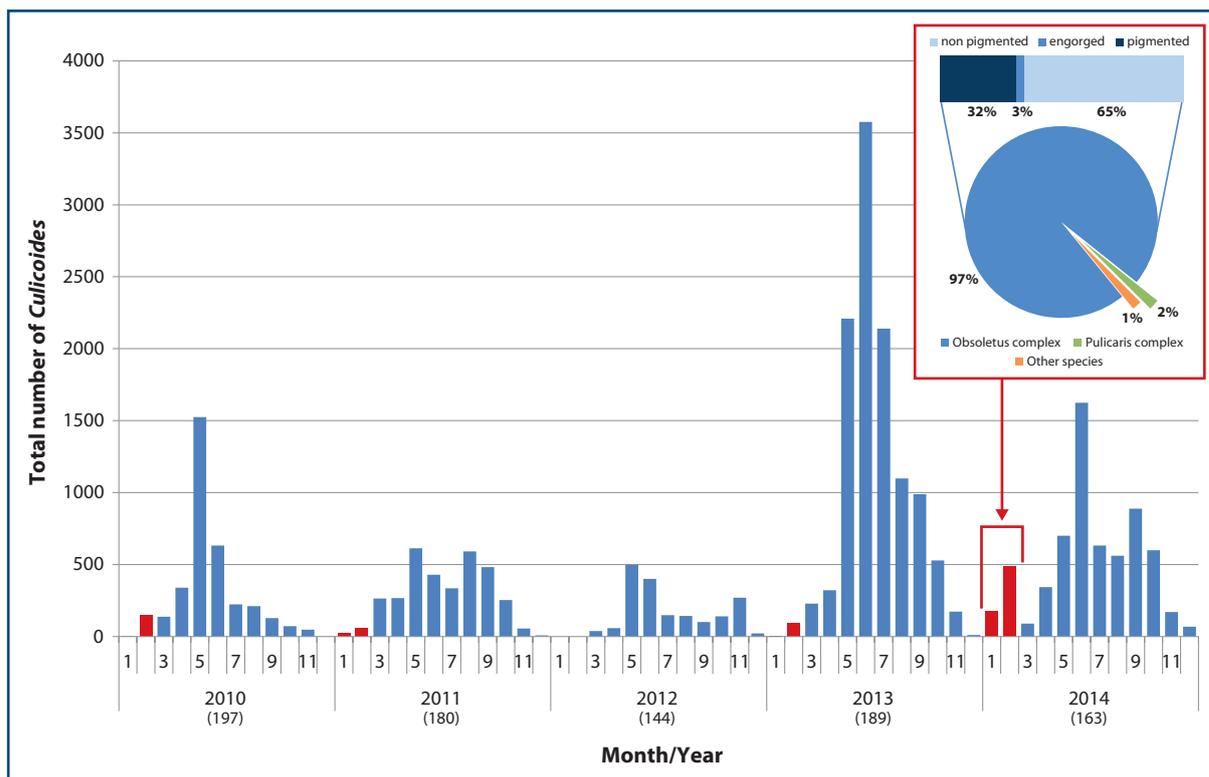
<sup>1</sup> [http://cib.knmi.nl/mediawiki/index.php/2014\\_warmest\\_year\\_on\\_record\\_in\\_Europe](http://cib.knmi.nl/mediawiki/index.php/2014_warmest_year_on_record_in_Europe).



**Figure 5.** Bluetongue virus serotype 1 circulation in 2013



**Figure 6.** Bluetongue virus serotype 1 circulation between end-December 2013 – March 2014 and permanent collection sites (green stars).



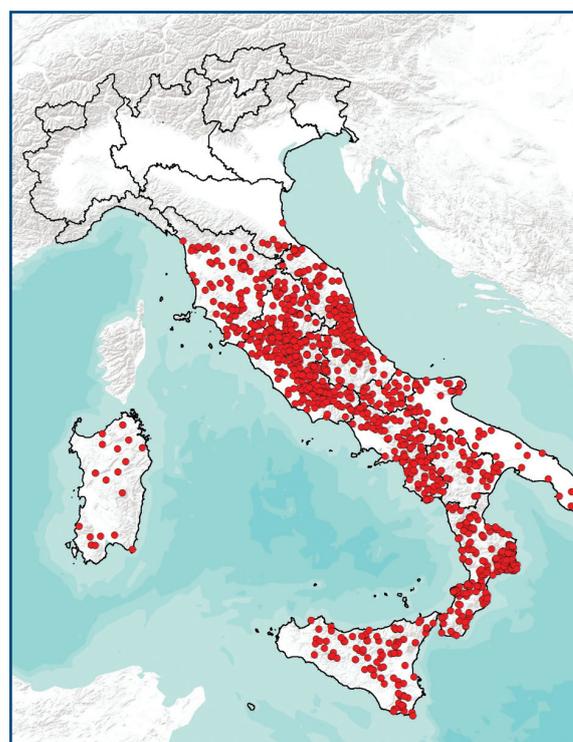
**Figure 7.** Seasonal distribution of the total number of *Culicoides* in the 4 permanent collection sites of the ‘overwintering’ area from 2010 to 2014 and the number of collections per year. Detailed information of the January-February 2014 collections is provided in the pie-chart .

These two factors (high rain and mild winter) might have contributed to the increasing of local *Culicoides* populations and to the survival of adult females of the *Obsoletus* complex possibly harboring the virus. This hypothesis is actually confirmed by the analysis of the traps permanently operating in the ‘overwintering’ area which showed higher values of monthly abundance of *Culicoides* spp. in January and February 2014, in comparison to the previous years (Figure 7).

The active *Culicoides* collected during the winter, were mainly identified as *Obsoletus* complex, including pigmented, engorged and unpigmented females (Figure 7). This finding indicates that the population was active and feeding on animals, possibly transmitting BTV. Under laboratory conditions, the *Obsoletus* complex showed a long life span (up to 104 days) when kept at a temperature between 17 and 25 °C, even without any blood meal (Goffredo et al. 2016), this again supporting the possible overwintering of BTV.

In summer 2014, the epidemic massively spread and by December 2014 all the regions in Central and Southern Italy were affected by BTV-1 (Figure 8). Summer 2014 was characterized by lower temperatures (especially for the maximum values) and higher precipitations and these two factors might have contributed to create a more suitable environment for *Obsoletus* complex that is able

to reach great abundance in areas characterized by higher elevation (an average of 587 m.), cooler temperature (minimum temperature between May



**Figure 8.** BTV1 distribution during the 2014 epidemics.

and November = 14.2°C) and with more vegetation in comparison with *C. imicola* (Conte *et al.* 2007). In addition, the BTV-1 strain responsible for the extensive spread in 2014 in Italy, was able to affect multiple species of *Culicoides*: besides *C. imicola* and the species of the *Obsoletus* complex, other species were found infected by BTV-1 (i.e. *Culicoides dewulfi*, *C. pulicaris*, *C. newsteadi*, and *C. punctatus*) (Goffredo *et al.* 2015).

The heavy circulation of BTV-1 and the severity of clinical outbreaks recorded in the regions of the Central Italy have a number of 'open questions' that are currently under investigation and several hypotheses have been proposed to explain the unusual spread of the disease. A specific adaptation

of the BTV-1 strain responsible for the 2014 outbreaks to the vector population of the areas and/or an improved vector capacity have been suggested. In this regard, multidisciplinary studies are needed to verify how climate could influence the adaptation mechanisms or the vector behaviors and affect the final vector capacity.

Specific genetic characteristics of the virus and vectors could also be considered and further molecular studies are ongoing in the attempt of investigating the existence of particular characteristics of the BTV-1 strain involved in the Italian 2014 epidemic and the *Culicoides* vector population of the affected areas.

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