# PREVALENCE OF BOVINE HERPESVIRUS 1 (BHV-1) INFECTION IN HUNGARIAN CATTLE HERDS

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Hungarian cattle herds were surveyed for bovine herpesvirus 1 (BHV-1) infection by ELISA of milk and serum samples. In 1993, 75% of the large cattle herds (consisting of more than 50 cattle) and all small herds (small-scale producers' stocks), while in 1997 90% of the small herds were included in the survey. In the case of large herds, 79.3% of the herds and 64.1% of the samples tested were found to be positive. Of the small herds, 13.5% and 15.7% tested positive in 1993 and 1997, respectively. The majority of large herds were Holstein-Friesian dairy stocks. Small herds with an infection rate markedly exceeding the average were found in those counties where the small herds had been in close contact with the large-scale farms, or where new herds were established by using animals of uncontrolled infectious bovine rhinotracheitis (IBR) status originating from large farms. Attention is called to the importance of maintaining the IBR-free status of small herds that constitute one-third of the Hungarian cattle population.

**Key words:** Bovine herpesvirus 1 (BHV-1), cattle, ELISA, serum, milk, prevalence, Hungary

The different disease entities caused by bovine herpesvirus 1 (BHV-1) cause substantial economic losses in the cattle herds. The disease occurs throughout the world (Straub, 1990). Since its appearance, serological tests for the detection of antibodies to BHV-1 have been performed in numerous countries in order to confirm BHV-1 infection or to elucidate the aetiology of diseases thought to be of viral origin (Msolla et al., 1981; Frost and Wagner, 1982; Albrecht et al., 1985; Edwards, 1988; Ackermann et al., 1989; Enders, 1989; Durham and Hassard, 1990; Niewöhner, 1990; Tanyi et al., 1990; Behymer et al., 1991; Hartman et al., 1997). Those surveys, however, were limited to relatively small administrative units and did not cover the entire cattle population of the countries concerned.

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Certain countries have embarked on eradication programmes either to diminish the economic losses (Ackermann et al., 1989, 1990) or to remove barriers to trade (Forschner et al., 1986). Herds (and, in some cases, countries) having completed the eradication programme may import animals exclusively herds that have a free status. A certain proportion of Hungarian breeding cattle are sold on markets where these provisions are in force. Apart from an earlier serological survey of smaller scale (Tanyi et al., 1990), no data were available on the BHV-1 infection status of the Hungarian cattle herds. Surveying the entire cattle population of Hungary for BHV-1 infection was necessitated by measures destined to maintain the exportability of bovine animals.

With these objectives in view, in 1993 75% of the large cattle herds (consisting of more than 50 cattle) and all small herds, while in 1997 90% of the small herds were surveyed for the prevalence of BHV-1 infection. The results are reported in this paper.

## Materials and methods

*Samples tested*

In both the large and the small cattle herds, milk and blood samples were tested exclusively from animals over 24 months of age. [Samples are submitted to the diagnostic institutes for qualification of the herds for enzootic bovine leukosis (EBL) status.]

*Small herds.* During the survey conducted in 1992–1993, milk samples (100 ml) taken by the attending veterinarian from the can were submitted from the dairy herds and individual blood samples (10 ml) from the beef herds. Samples were submitted from all herds. In 1996–1997, only individual blood samples were submitted to the institutes from all herds. At that time, samples were sent exclusively from herds that had been declared free of EBL in 1993 (these herds represented about 90% of the total population of small cattle herds).

*Large herds.* Depending on the milking equipment used (milking in herringbone milking parlours with 8 stalls or multiples of 8 stalls), the attending veterinarian pooled identical volumes of milk samples taken from 8 cows and submitted 100 ml of that pooled sample for testing. From non-dairy stocks and from dry cows individual blood samples were submitted. According to the current EBL status, only about 75% of the large herds were included in the survey.

*Sample processing*

*Serum samples.* In the case of blood samples submitted from the large herds, aliquots of 10 sera were pooled in the laboratory to obtain the samples to be tested. In the case of the small farms, test samples were obtained by pooling

aliquots of only those serum samples which originated from animals kept in the same yard. In most cases, samples of less than 10 animals were pooled to obtain a single sample.

*Milk samples.* The milk samples were considered processed for testing.

*ELISA*

The ELISA procedure was performed with a slight modification of the method described earlier (Tekes et al., 1991; Tekes and Juhász, 1993). In brief: milk samples were diluted 1:2 in a diluent, while blood samples were diluted 1:150 (for one serum). Subsequently 0.2 ml aliquots of the samples were measured into two neighbouring wells of the ELISA plate, one of which had been sensitised with antigen while the other one with control antigen. The sera were incubated on the plate at 37 °C for 90 min, washed three times, then 0.2 ml of monoclonal anti-bovine IgG-horseradish peroxidase (HRPO) conjugate was added to each well, and the plates were again incubated at 37 °C for 90 min. Then the plates were washed and 0.1 ml 3,3',5,5'-tetramethyl benzidine (TMB, Sigma) chromogen was added to the wells. After 10 min, the reaction was stopped by the addition of 0.05 ml of 4 N sulphuric acid. The results were read using a Multiscan R Plus photometer (Labsystem, Finland). Evaluation was done with an own-developed software as described previously.

## Results

The results obtained in the survey of the large herds are shown, broken down by county, in Table 1. It can be seen that 79.3% of the large herds surveyed and 64.1% of the samples tested proved to be positive. The results obtained for the small herds are presented in Table 2: 13.5% and 15.7% of the herds tested positive in 1993 and 1997, respectively.

## Discussion

From the 1960s, the structure of the Hungarian cattle husbandry was characterised by small household ('backyard') stocks consisting of 1–2 cows and large-scale cattle herds of 300–1500 cows. The government policy that took shape at the beginning of the 1990s intended to restructure the agriculture by establishing Western-type family farms with 20–40 hectares of land and 20–

80 cows. Using animals from the large herds[*] seemed to be the most feasible solution to attain that goal. However, the diseases that these stocks would be affected with were not at all negligible from the animal health point of view regarding the future disease status and sales potential of the herds concerned.

**Table 1**

IBR infection of large herds by county in 1993

| County | No. of large herds surveyed | Positive herds | | No. of test samples | Positive samples | |
|---|---|---|---|---|---|---|
| | | number | % | | number | % |
| Bács | 56 | 47 | 83.9 | 12409 | 5387 | 43.4 |
| Baranya | 9 | 7 | 77.8 | 350 | 280 | 80 |
| Békés | 57 | 51 | 89.5 | 2742 | 1526 | 55.6 |
| Borsod | 32 | 24 | 75 | 781 | 532 | 68.1 |
| Csongrád | 43 | 42 | 97.7 | 997 | 920 | 92.3 |
| Fejér | 47 | 42 | 89.4 | 9743 | 8057 | 82.7 |
| Győr | 35 | 8 | 22.8 | 1307 | 29 | 2.2 |
| Hajdú | 53 | 45 | 60 | 4348 | 2935 | 67.5 |
| Heves | 36 | 28 | 77.8 | 941 | 716 | 76.1 |
| Komárom | 14 | 13 | 92.9 | 1202 | 940 | 78.2 |
| Nógrád | 47 | 44 | 93.6 | 9927 | 7859 | 79.2 |
| Pest | 33 | 31 | 94 | 8170 | 7077 | 86.6 |
| Somogy | 59 | 43 | 79.9 | 1413 | 1029 | 72.8 |
| Szabolcs | 67 | 59 | 88 | 7020 | 3196 | 45.5 |
| Szolnok | 30 | 29 | 97 | 1503 | 941 | 62.6 |
| Tolna | 4 | 2 | 50 | 50 | 24 | 48 |
| Vas | 53 | 29 | 54.7 | 2236 | 154 | 6.9 |
| Veszprém | 12 | 11 | 91.7 | 3078 | 2686 | 87.3 |
| Zala | 48 | 28 | 60.4 | 1924 | 543 | 28.2 |
| Capital Budapest | 1 | 1 | 100 | 384 | 364 | 94.8 |
| Total | 736 | 584 | 79.3 | 70525 | 45195 | 64.1 |

Earlier information on the BHV-1 infection status of large cattle herds was available from certain regions of Hungary (Mocsári et al., 1973; Tanyi et al., 1990) but not from the country as a whole. As a result of the vaccination programmes applied (Bartha et al., 1973), the diseases that had assumed considerable proportions in earlier years ceased to exist by the mid-1980s or manifested themselves in negligible clinical symptoms only. Simultaneously with

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[*] Animal holding with a large stock of animals: holdings with more than 50 head of cattle, as defined in Appendix no. 1 of Decree no. 41/1997 (V.28.) FM of the Ministry of Agriculture on the issue of the Animal Health Code

that, vaccinations were mostly discontinued in the large herds, while in the small herds no IBR vaccine was used at all. Recently the infection of herds was not considered important, apart from those infections which were subject to export restrictions.

**Table 2**

IBR infection of small herds by county in 1993 and in 1997

| County | 1993 | | | 1997 | | |
|---|---|---|---|---|---|---|
| | No. of small herds surveyed | Positive herds | | No. of small herds surveyed | Positive herds | |
| | | number | % | | number | % |
| Bács | 5777 | 830 | 14.4 | 4548 | 752 | 16.4 |
| Baranya | 2639 | 285 | 10.8 | 1182 | 162 | 13.7 |
| Békés | 3438 | 556 | 16.2 | 3072 | 520 | 16.9 |
| Borsod | 5506 | 328 | 5.9 | 4397 | 609 | 13.8 |
| Csongrád | 3739 | 465 | 12.4 | 3278 | 396 | 12.0 |
| Fejér | 1237 | 340 | 27.5 | 724 | 167 | 23.1 |
| Győr | 4023 | 134 | 3.4 | 2486 | 357 | 14.4 |
| Hajdú | 8373 | 1343 | 16 | 6597 | 711 | 10.8 |
| Heves | 1133 | 70 | 6.2 | 1025 | 234 | 22.8 |
| Komárom | 390 | 137 | 35.1 | 419 | 81 | 19.3 |
| Nógrád | 721 | 209 | 29 | 633 | 134 | 21.2 |
| Pest | 2383 | 677 | 28.4 | 2189 | 523 | 23.9 |
| Somogy | 2239 | 291 | 13 | 833 | 125 | 15.0 |
| Szabolcs | 9008 | 1124 | 12.5 | 5340 | 840 | 15.7 |
| Szolnok | 3282 | 535 | 16.3 | 3358 | 646 | 19.2 |
| Tolna | 1559 | 129 | 8.3 | 979 | 268 | 27.3 |
| Vas | 3356 | 210 | 6.3 | 2255 | 269 | 11.9 |
| Veszprém | 1681 | 451 | 26.8 | 1154 | 260 | 22.5 |
| Zala | 2855 | 431 | 15.1 | 1395 | 157 | 11.3 |
| Capital Budapest | 34 | 17 | 50 | 35 | 10 | 28.6 |
| Total | 63373 | 8562 | 13.5 | 45899 | 7221 | 15.7 |

During the survey conducted in 1992–1993, 79.3% of the large herds were found to be positive for infection. Most of these herds consisted of dairy cows of the Holstein-Friesian breed. The results are not surprising in the light of earlier surveys conducted elsewhere, all of which showed a high prevalence of BHV-1 infection in Holstein-Friesian cattle or herds (Msolla et al., 1981; Bölle et al., 1990; Durham and Hassard, 1990). The infection rate was found to rise in direct ratio to herd size (Enders, 1989). Still, it was surprising to see that the infection rate of large cattle herds of counties Győr, Vas and Zala was markedly lower

than that of the other counties, although these counties did not differ from the others in terms of breed structure and herd size.

About one-third of the Hungarian cattle population (120,000–130,000 cows) can be found in small herds. The herd infection rate of 13.5%, found in 1993, fully conformed to our earlier assumptions. The differences between counties in infection rate can be explained by the close, direct contact with large herds (transfer of animals, purchase of Holstein-Friesian cattle) in certain counties (Fejér, Komárom, Pest, Veszprém). The survey conducted in 1997 revealed that 15.7% of the small herds were infected with BHV-1. Although the increase that took place in infection rate during the four years that had elapsed since the previous survey was not substantial on national scale, the unfavourable changes seen in certain counties (Borsod, Győr, Heves, Tolna) call attention to the gradually increasing prevalence of BHV-1 infection in small herds as a factor to be considered when establishing new herds. To counter that trend, closer attention should be paid to preserving the BHV-1 free status of small herds constituting one-third of the entire cattle population. When establishing new herds, the IBR-free status of cattle to be transferred to such herds should be required. In order to have sufficient numbers of BHV-1 free cattle to be used for the establishment of new herds, and to maintain our cattle exports on the previous level, the eradication of BHV-1 from the large herds would have to be started immediately.

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