

The prevalence of anti-*Echinococcus* antibodies in the North-Western part of Romania

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Abstract. The serological testing is important for diagnosis of patients and for knowledge the distribution of the parasite in different territories. To assess the level of the infection in the healthy population and to establish prevalence of anti- *Echinococcus* antibodies, we performed a serological screening in 683 people without any symptoms of disease. Six counties from Transylvania were investigated: Bihor (92), Satu-Mare (119), Salaj (141), Bistrița-Năsăud (84), Cluj (138) and Alba (109). The prevalence of the hydatidosis in the screened population was 2.59%. Between all categories, the country side and female groups has a significant higher seropositivity level. No significant difference was found between age groups.

Keywords: anti-*Echinococcus* antibodies; Serological screening; Seroepidemiology; Human cystic echinococcosis; ELISA.

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Introduction

Human cystic echinococcosis caused by the larval stage of *Echinococcus granulosus* is a major zoonosis, having variable global distribution and geographical incidence (Eckert and Deplazes, 2004).

The serologic screening helps us understanding the distribution of the parasite in different areas in order to initiate certain surveillance programs and controlling the disease. Starting from these grounds and from the fact that the real prevalence of the infection is in accordance with the “iceberg principle”

(i.e. obviously higher), we carried out screening to evaluate the presence of anti-hydatid antibodies in samples of an apparently healthy population.

Materials and methods

We underlined the presences of *E. granulosus* parasite in apparently healthy population by a serologic study carried out on 683 representative samples from the 6 counties of Transylvania: Bihor (92), Satu-Mare (119), Sălaj (141), Bistrița-Năsăud (84), Cluj (138) and Alba (109). We chose samples from the North-Western part of the country due to the

fact that this area was considered endemic for our country according to some prospective serologic studies carried out by Coroiu et al. (1995a; b).

We used the immunoenzymatic test (ELISA) of IgG antibodies (NovaTec kits). The sensibility and specificity of ELISA test for these kits is >95%. The working dilution of the sample was 1/100. The photometric measurement was done at 450 nm. The results were expressed in NovaTec kit units (NTU). Values below 9 NTU were considered negative, those between 9 and 11 NTU were labeled as not relevant while those over 11 NTU were considered positive.

We used the student test ("t" test) for the comparison between the two percentages as statistical calculation method. The Bayes' theorem was used in order to determine the positive predictive value of the test applied to the chosen sample.

Results and discussion

We analyzed the distribution of anti-hydatid antibodies depending on the origin of seropositive persons, on sexes and age categories, in order to establish the hydatid infection groups of risk. We draw up a prediction of the disease by expressing the positive predictive value.

Regional situation of serologic investigations

The graphical representation of the hydatid prevalence established by the tests and the prediction of hydatid disease depending on the studied categories of population is shown in figure 1. The positive probability value obtained for the whole investigated area demonstrated that the positive serologic results are in accordance with the 50.6% presences of cystic echinococcosis in human bodies. By applying the probability values to the level of the general positivity, it became 2.59% instead of 5.12%, level to which we preedict the presence of cystic echinococcosis in the population of the investigated area. The results of our investigations show the fact that a percentage of the investigated population is carrier of anti-hydatid antibodies, but there are not hydatid cysts in their body. The fact that

the presence of hydatid antibodies is not similar to the presence of hydatid cyst, even in asymptomatic cases, was demonstrated by several authors (Barbieri et al., 1994; Arda et al. 2009) by ultrasonography.

The zonal frequency of hydatid antibodies and of predicted disease risk

By the graphical representation of hydatid antibodies and of predicted disease level in the 6 counties (figure 2), we noticed variations of the seropositivity and of the predicted disease risk from one area to another. The prediction regarding the cystic echinococcosis is situated among the minimum of 1.96% in Sălaj County, 2.02% in Cluj County, 3.19% in Satu-Mare County and 4.63% in Bihor County.

In figure 2, we may notice the delimitation of 2 different seropositivity and implicitly hydatid disease areas. Thus we have an area in which seropositivity and the prediction of hydatid disease risk are higher, such as the North-Western extreme areas, Satu-Mare and Bihor counties where we have 6.63% seropositivity and 3.81% prediction of disease. Then we may notice an area with lower values in the central part of the North-Western area, including Bistrița-Năsăud and Alba counties, Cluj and Sălaj counties, where we have 4.44% seropositivity and 0.56% prediction of hydatid disease. Based on these grounds we may say that in the asymptomatic population of the studied area there are gradations in what regards the level of infection with *E. granulosus*.

Statistical analyses revealed significant differences between the first and the second areas as regards prevalence in asymptomatic population ($p < 0.01$).

There are many causes of the division into zones, but first of all we have to take into consideration the interactions between the parasite and the host and the characteristics of each in part. The variations noticed in the studied area are related to the local characteristics of animal breeding and to the characteristics of the host and of the parasite that develops in the territory.

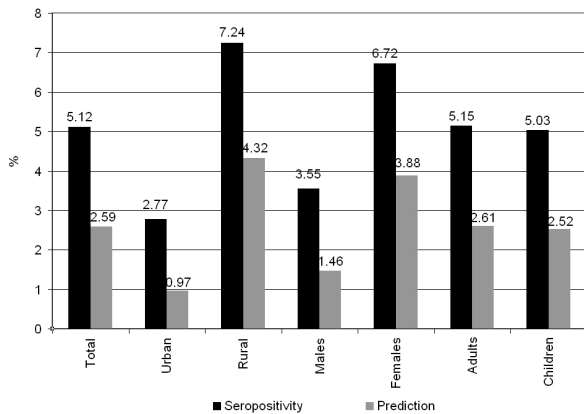


Figure 1. Regional situation of the serologic tests

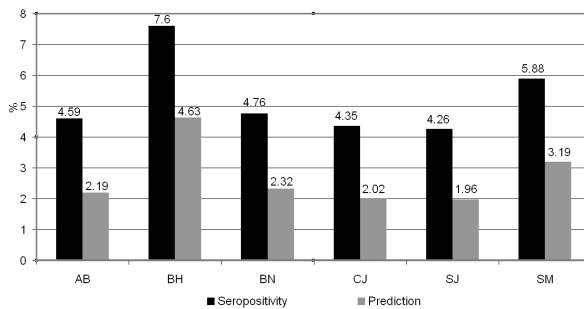


Figure 2. Zonal prevalence of anti-hydatid antibodies and of predicted disease

Even with these small but constant variations, anti-hydatid antibodies are present in the investigated area. This fact proves the presence of the parasite and the existence of perpetuating conditions of its biologic cycle, with direct consequences on its transmission to the humans.

Serologic study of cystic echinococcosis in urban and rural environments

By comparing hydatid antibodies depending on the origin of the subjects, we may notice that their level is higher in rural than in urban environment, both at each county level as well as overall regional level. In the whole area (figure 3) we noticed a 7.24% frequency of antibodies in rural population in comparison with 2.77% in urban population ($p < 0.01$). The risk of infection is 2.6 higher in rural than in urban environment.

The possibility of acquiring the infection is significantly higher under rural life conditions. The predicted presence of the disease in rural environment (4.32%) is 4.4 higher than in urban environment (0.97%). This situation is maintained by the insufficient knowledge about the transmission of the parasite, by the human behavior when feeding dogs and by the removal of infested organs resulted from slaughtered animals. At county level we may notice variations of Rural/Urban (R/U) report, by taking into consideration the level of antibodies in the population and the probability of clinical infection. This report is smaller in Alba (R/U=2.4) and in Bistrița-Năsăud (R/U=2.1) counties, and higher in Cluj (R/U=18.2) and Sălaj (R/U=8.8).

These variations appeared on the background of social-economic changes in the area. We consider that the presence of the parasite in human population is higher in the rural environment. Even if there is a higher risk of infection in the rural environment, we cannot exclude the possibility of parasite transmission in urban environment, especially due to the massive presence of feral dogs. They represent a real danger in the pollution of the environment with infective eggs (Shambesh et al., 1999; Baharsefat et al., 2007; Mirzanejadasl et al., 2008).

Serologic study of cystic echinococcosis depending on sexes

By comparing anti-hydatid antibodies depending on sexes, we may notice that their level is significantly ($p < 0.01$) higher in women than in men, on the whole studied area. In the whole area we may notice a 6.72% frequency of anti-hydatid antibodies in females than 3.55% in males (figure 4). The risk of infection is 1.8 higher in females than in males. The female population is the most exposed category to the risk of infection. The 3.88% disease prediction in females is 2.6 higher than the 1.46% prediction in males.

The most significant differences are in Bihor (F/M=4.9) and Satu-Mare (F/M=6.4) counties and less in Cluj (F/M=1.2), Sălaj (F/M=1.4) and Bistrița-Năsăud (F/M=1.4) counties.

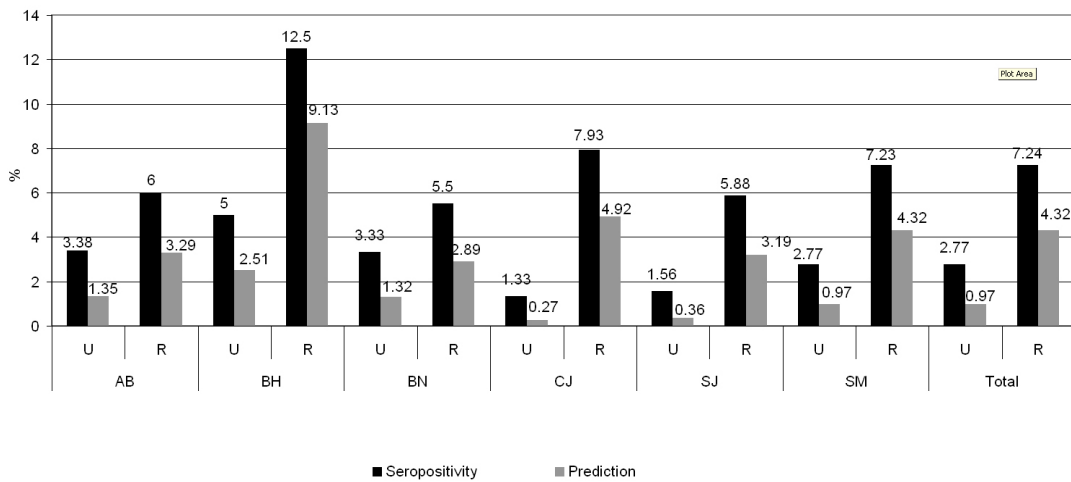


Figure 3. Serologic results for anti-*Echinococcus* antibodies in urban (U) and rural (R) environments

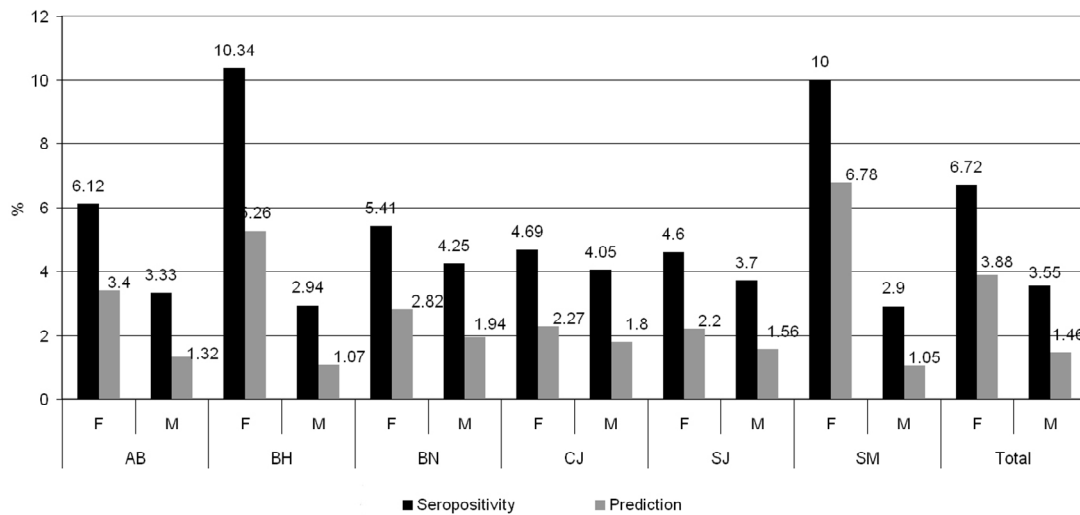


Figure 4. Serologic results for anti-*Echinococcus* antibodies in females (F) and males (M)

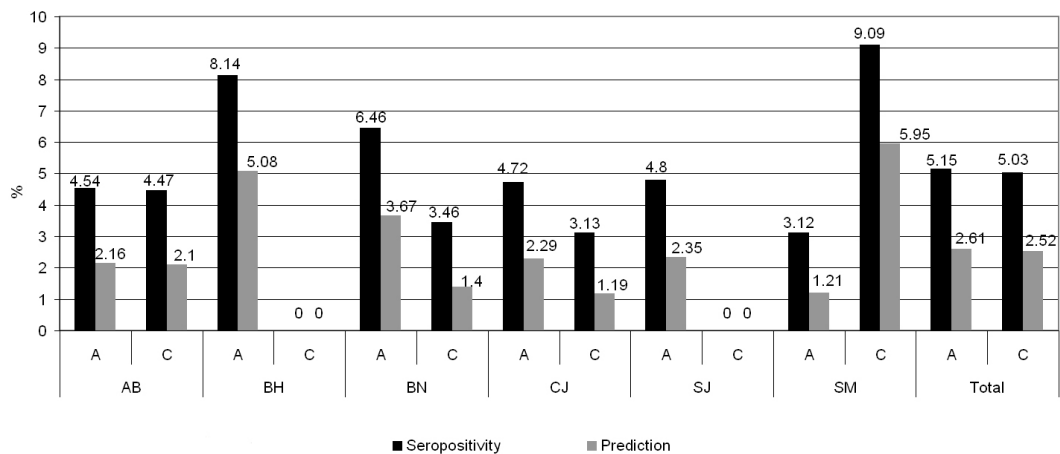


Figure 5. Serologic results for anti-*Echinococcus* antibodies in adults (A) and children (C)

Serologic study of cystic echinococcosis in children and adults

By comparing hydatid antibodies in adult and child population we found no statistical differences: 2.61% prevalence in adults; 2.52% in children (figure 5). However, the investigated groups were not homogenous in number. Only in Satu Mare County the prediction for cystic echinococcosis was significantly higher in children (5.95%) compared to adults (1.21%). Thus, infantile population must be considered for further epidemiologic studies.

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