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IVTH INTERNATIONAL SYMPOSIUM ON RICKETTSIAE AND RICKETTSIAL DISEASES

PREFACE

The IVth International Symposium on Rickettsiae and Rickettsial Diseases organized by the Institute of Virology, Slovak Academy of Sciences, Bratislava was held in Piešťany Spa, October 1-6, 1990.

The Symposium was attended by 101 participants from 23 countries of all continents (Australia, Japan, People's Republic of China, Afghanistan, Israel, Egypt, Zimbabwe, U.S.A., Scotland, Sweden, Finland, Portugal, Spain, France, Italy, Federal Republic of Germany, Switzerland, Austria, Yugoslavia, Bulgaria, Poland, U.S.S.R., and Czech and Slovak Federal Republic) including a representative of the World Health Organization. They represented both scientists involved in basic rickettsiological research and public health workers from those countries in which rickettsial diseases pose or may pose a serious public health problem.

The Scientific Programme of the Symposium was divided into 7 sessions as follows: 1. ultrastructure and chemical structure of rickettsiae, 2. molecular biology and genetics of rickettsiae, 3. pathogenesis of and immunity in rickettsial infections, 4. ecology of rickettsiae and epidemiology of rickettsial disease, 5. ehrlichiae and ehrlichiosis, 6. diagnosis of rickettsial diseases, 7. control and therapy of rickettsial diseases. In these sessions 14 introductory lectures and 64 short communications were delivered. In addition, there were 26 presentations in a separate Poster Session.

Because the introductory lectures are published in this issue of the journal, we briefly mention the contributions presented as short communications and in the Poster Session, which all should appear as the Proceedings of the Symposium published by VEDA, Publishing House of the Slovak Academy of Sciences, Bratislava in the first half of 1991.

All three papers on the Ultrastructure of Rickettsiae dealt with *Coxiella burnetii:* one on the sporogenic differentiation of *C. burnetii* during its developmental cycle within the affected phagolysosomes (McCaul et al., Australia, U.S.A.), another on the ultrastructure of *C. burnetii* in mice during acute infection and vaccinal process (Popov et al., U.S.S.R.) and the last one on the interaction between *Rickettsiella phytoseiuli* and *C. burnetii* in *Dermacentor reticulatus* ticks. (Šudăkova' and Rèhácèk, C.S.F.R.).

Papers on the Chemical Structure of Rickettsiae can be divided into two groups: three on typhus rickettsiae and four on *C. burnetii.* In the former, the surface array protein antigens as the dominant protein immunogens of typhus rickettsiae differentiating species of this serological group were analyzed structurally and immunologically (Ching et al., U.S.A.), while the remaining two papers by Balayeya et al. (U.S.S.R.) studied the protein antigens and the isolation and characterization of a 100 kD protein of *Rickettsia prowazekii* strains with different virulence. In the latter, the structural heterogeneity of the polysaccharide component of *C. burnetii* lipopolysaccharide was presented (Toman and Kazár, C.S.F.R.), the immunochemical properties of *C. burnetii* strains Nine Mile and Priscilla, causing acute and chronic infections, respectively, were compared (Lukáčová et al., C.S.F.R. and F.R.G.), monoclonal antibodies to *C. burnetii* were characterized (Sekeyová et al., C.S.F.R.), and proteins and lipopolysaccharide of Chinese *C. burnetii* strains were analyzed by SDS - Page and immunoblot with monoclonal antibodies (presented by Yu Shu-rong et al., People's Republic of China).

In the Session on the Molecular Biology and Genetics of Rickettsiae, 7 short communications were delivered. A study on the nucleotide metabolism of *R. prowazekii* (Winkler et al., U.S.A.) revealed the transport of UXP, but not the transport of thymidylates, thymidine, uracil, uridine or dUMP, from the host cell's cytoplasm by the rickettsiae. The polymerase chain reaction technique has been used for genotypic identification of rickettsiae (Regnery, U.S.A), for the isolation of the *R. prowazekii* gyrA gene (Waite and Wood, U.S.A.) and for the identification of spotted fever group rickettsiae tick isolates (Drancourt et al., France, U.S.A.). In further studies on *C. burnetii,* strain-specific plasmid sequences were analyzed (Minnick et al., U.S.A.), plasmid size of eight new isolates was determined (Stein et al., France) and behaviour of the strains causing acute infection (with QpH1-type plasmids) and chronic infection (plasmidless or with QpRS-type plasmids) was compared in L929 cell cultures (Baca et al., U.S.A.). Chronic *C. burnetii* isolates with QpRS-type plasmids took longer to infect cells, exhibited a higher degree of vacuolation and displayed significantly less *C. burnetii*-specific antigen on the host cell membrane.

The Session on the Pathogenesis of and Immunity in Rickettsial Infections consisted of 11 papers. The first paper is on the inflammatory infiltrates in intradermal *R. prowazekii* reinfection in man following the recruitment of T lymphocyte types which are a rich source of gama interferon and can be cytotoxic for rickettsiae-infected cells (Dumler and Wisseman, U.S.A.).

Histopathology and immunopathology of skin biopsy specimens in patients with Mediterranean spotted fever was presented and proved to be a rapid and sensitive method of identification of rickettsiae in skin lesions (Dujella et al., Yugoslavia).

Two papers analyzed the mechanism of hyponatremia and reported mixed cryoglobulinemia in Mediterranean spotted fever (Herrero Herrero et al., Spain). In another study, the role of tissue factor-like procoagulant activity produced by human leukocytes in the pathogenesis of *C. burnetii* endocarditis was suggested (Miragliotta, Italy). Sexual transmission of *C. burnetii* infection was investigated and the negative effects of *C. burnetii* infection in male mice on their offspring was demonstrated (Kruszewska and Tylewska-Wierzbanowska, Poland). Interferon-resistant strains of *R. prowazekii* have been demonstrated and analyzed (Turco and Winkler, U.S.A.). The reduction of *Rickettsia conorii* growth in permissive HEp-2 cells caused by their incubation with monocyte-derived macrophages was shown to be associated with higher tumour necrosis factor and interleukin-1 secretion (Manor, Israel). Increased resistance to *Salmonella typhimurium* infection was demonstrated in mice immunized with *C. burnetii* antigen (Tokarevich et al., U.S.S.R.). The remaining two papers analyzed antibody response to *C. burnetii* in mice by immunoblotting (Novák et al., C.S.F.R.; Thiele et al., F.R.G.).

The greatest number of papers, a total of 17, were presented in the Session on Ecology of Rickettsiae and Epidemiology of Rickettsial Diseases. New data on the establishment of *Rickettsia typhi* infection in native populations of rodents and other small and medium-sized mammals may gradually expand the distribution of murine typhus into suburban and rural areas of the world (Azad, U.S.A.). Direct immunofluorescence antibody test, polymerase chain reaction, tissue culture plaque assay and animal inoculation assays were used for identitification of spotted fever group rickettsiae in ticks from the North Sinai, Egypt (El Dessouky et al., Egypt, Israel, U.S.A.). Three papers were devoted to *Rickettsia japonica*, one on its isolation and identification (Uchida et al., Japan), another on its analysis with species-specific monoclonal antibodies (Uchiyama et al., Japan, U.S.A.), and the last one on the clinical features of the disease it causes - Japanese spotted fever (Mahara and Suto, Japan). A study on the occurrence of unknown fever caused by rickettsiae belonging to the spotted fever group was reported from the Astrakhan's region of U.S.S.R. (Tarasevich et al., U.S.S.R., France). *R. conorii* was detected in the wild environment of the province of Salamanca in Spain (Ruiz Beltran et al., Spain) and *R. typhi* and *R. conorii* were isolated in rodents trapped in the region of Lazio in Italy (Lillini et al., Italy). Epidemiological studies on the occurrence of tick-bite fever (*R. conorii* infection) in Zimbabwe (Kelly and Mason, Zimbabwe); Mediterranean spotted fever in Sicily (Scarlatta et al., Italy); and Q fever in Spain (Hellín, Spain), Southern Bohemia (Vošta et al., C.S.F.R.) and in Leszno district in Poland (Tylewska-Wierzbanowka et al., Poland) were reported. Rickettsiae and rickettsia-like organisms detected in *Rhipicephalus sanguineus* ticks collected in Portugal were analyzed (Bacellar et al., Portugal, C.S.F.R.). Extensive examination of different tick species for infestation with *C. burnetii* in Slovakia resulted in the isolation of 10 strains from 5 tick species (Řeháček et al., C.S.F.R.). The imunofluorescent hemocyte test using monoclonal antibodies revealed the infestation of ixodid ticks by *C. burnetii* and spotted fever group rickettsiae in Bulgaria (Serbezov et al., Bulgaria). Finally, some practical problems of animal coxiellosis and the dissemination of *C. burnetii* from a primary focus of infection were discussed (Aitken, Scotland).

In the Session on Diagnosis of Rickettsial Diseases, one presentation dealt with DNA probes for the rapid detection of *C. burnetii* and for differentiation between *C. burnetii* strains associated with chronic and acute disease (Frazier et al., U.S.A.) while the remaining 6 papers dealt with serological diagnosis. The cross-reactivity between rickettsiae and *Proteus* antigens in the Weil-Felix test with sera from rickettsial diseases patients was analyzed by ELISA and immunoperoxidase test suggesting that the reactivity of patients sera (with tsutsugamushi disease and Japanese spotted fever) in the Weil-Felix test is dependent on the IgM antibody (Amano et al., Japan). Selective reactivity of rabbit antibodies to *R. typhi*-erythrocyte substance was revealed by immunoelectron microscopy and immunoblot test (Hechemy et al., U.S.A.). A summary of ten years of experience on the diagnosis of rickettsial diseases using indirect imunoperoxidase methods indicated that these may serve as rapid and accurate serologic diagnostic procedures (Suto, Japan). Immunoblot technique did not show any difference between *R. conorii* and Israeli spotted fever rickettsiae antigens (Hanuka et al., Israel). No detectable difference was found by immunoblotting between symptomatic vs. asymptomatic spotted fever group rickettsiae cases (Sarov et al., Israel). A comparison of 4 serologic tests for diagnosis of Mediterranean spotted fever demonstrated the superiority of microimmunofluorescence and immunoperoxidase tests over complement fixation and Latex agglutination tests (Novaković et al., Yugoslavia).

Of 6 papers presented in the Session on Control and Therapy of Rickettsial Diseases, 4 were connected with rickettsial vaccines and 2 with antibiotic therapy. In order to produce a subunit vaccine, the recognition of natural and synthetic peptide fragments of the

major surface protein protective antigen of typhus group rickettsiae by rickettsia-specific human T lymphocytes was analyzed (Churilla et al., U.S.A.). Cross-immunity between 4 different phase I *C. burnetii* strains was demonstrated in guinea pigs, indicating the possibility of the use of a monovalent corpuscular or soluble Q fever vaccine (Lesný et al., C.S.F.R.). In two papers (Gajdošová et al., C.S.F.R., Kováčová et al., C.S.F.R.), the immunogenicity of a Q fever chemovaccine was evaluated on human volunteers at different post-vaccination intervals. The lymphocyte transformation test was more sensitive for the detection of post-vaccination immunity than serological tests, of the latter group the ELISA and microimmunofluorescence tests were more sensitive than microagglutination and complement fixation tests. Alkalization of phagolysosome was demonstrated to increase the susceptibility of *C. burnetii* to doxycycline (Maurin et al., France). New quinolones were compared with other antibiotics in the treatment of Mediterranean spotted fever suggesting the use of ciprofloxacin for those cases in which the use of tetracyclines is not advisable (Ruiz Beltran and Herrero Herrero, Spain).

The session on Ehrlichiae and Ehrlichiosis was represented by 6 short communications. The distribution and clinical spectrum of human ehrlichioses in the United States from 1985 to 1989 was presented (Fishbein and Dawson, U.S.A.). Tick-borne fever was found to pose a significant disease problem in cattle in Switzerland (Liz et al., Switzerland). In two papers the effects of antibiotics on the phagosome-lysosome fusion of *Ehrlichia sennetsu*-infected cells (Brouqui and Raoult, France) and ehrlichiacidal mechanism by macrophage Ca^2 mobilization (Rikihisa and Park, U.S.A.) were studied. One paper was dedicated to the ehrlichial diagnosis, namely detection of *Ehrlichia risticii* using a gene probe and polymerase chain reaction amplification (Dutta et al., U.S.A.). The final paper presented the development of a cell line for continuous *in vitro* propagation of *Ehrlichia canis* (Holland and Ristic, U.S.A.).

There were 24 contributions presented in the Poster Session. Comparison of the ultrastructure of the cell envelopes of *C. burnetii* and *R. phytoseiuli* made it possible to differentiate between these two agents (Popov et al., U.S.S.R., C.S.F.R.).

DNA restriction endonuclease analysis was used for interspecies differentiation of typhus group rickettsiae (Artemiev et al., U.S.S.R.). Trasformation procedures were established for *Rochalimaea quintana* and pulse-field electrophoresis was employed to develop physical maps of chromosomal DNA of *Rochalimaea* spp. (Reschke et al., U.S.A.). This method was also used for the physical mapping of the *C. burnetii* genome (Frazier et al., U.S.A.). Evidence for the occurrence of domestic Q fever in Sweden was discussed in two papers, one described antibodies in the sera of sheep farmers and their families on Gotland Island by ELISA directed to a 60 Kd protein as analyzed by Western blotting (Norlander et al., Sweden), and the other isolation of *C. burnetii* from sheep placenta from this island (Kindmark et al., Sweden, U.S.A.). The comparison of proteins and lipopolysaccharides of *C. burnetii* isolates from ticks collected in Slovakia revealed several differences related to the localities in which the infectious ticks were collected rather than the tick species examined (Vavreková et al., C.S.F.R.). The level of antibody response to experimental infection with *C. burnetii* and spotted fever group rickettsiae was higher in wild rodents than in laboratory white mice, indicating that the former were more susceptible to the rickettsial species under study (Kocianová et al., C.S.F.R.). When comparing the antibody response in acute Q fever patients treated and untreated with antibiotics, it was found that antibodies to *C. burnetii* appeared later and in lower titres in the sera of antibiotic-treated patients (Seguljev et al., Yugoslavia). Analysis of the present status on Q fever in the province of Olsztyn, Poland, verified the presence of *C. burnetii* in the region with the absence of human Q fever cases (Anusz et al., Poland, C.S.F.R.). Additional data on the occurrence of tick-bite fever in Zimbabwe were presented (Kelly and Mason, Zimbabwe). The use of Western blotting in seroepidemiology to determine the kinetics of specific antibodies in Mediterranean spotted fever was suggested (Beati et al., France). Thousands of serum samples from three different regions of the U.S.S.R. were tested for the presence of antibodies to *R. prowazekii, Rickettsia sibirica* and *C. burnetii* to gather seroepidemiological data on the occurrence of rickettsioses in this country (Dodonov et al., U.S.S.R.). Two papers dealt with rickettsial infections in the North-Dalmatian subregion of Yugoslavia: one presenting the results of a seroepidemiological study among rural and urban populations (Dželalija et al., Yugoslavia), another describing the clinical spectrum of rickettsioses in a given area (Morović et al., Yugoslavia). A number of studies were conducted on the clinical manifestations of rickettsial infections and their treatment: a clinical study of Q fever patients in Spain (Hellín Sanz, Spain), a case report of pericarditis during Boutonneuse fever (Petrosillo et al., Italy), a very interesting case of tsutsugamushi disease probably infected in Africa (Suto et al., Japan), elevated serum adenosine deaminase level as a marker of acute Mediterranean spotted fever (Espejo et al., Spain), efficacy of pefloxacin in the treatment of Boutonneuse fever (Petrosillo et al., Italy), and a comparison of rifampin and doxycycline for Mediterranean spotted fever therapy (Bella et al., Spain). Antibiotic susceptibilities and possible resistance mechanisms of several *C. burnetii* isolates both from acute and chronic Q fever cases were analyzed (Baca and Yeaman, U.S.A.). The reactogenicity and immunogenicity of three Q fever chemovaccine preparations obtained under different conditions of trichloroacetic conditions were compared (Lesná et al., C.S.F.R.). Finally, the first verified cases of granulocytic ehrlichiosis in horses in Sweden were documented (Bjoersdorff et al., Sweden).

As can be seen from this review of the papers presented at the Symposium, practically all areas of research surrounding rickettsiology, both theoretical and practical, were covered. Of interest is the fact that several studies were carried out with international collaboration, which is obviously necessary in order to deal with some of the problems concerning not only rickettsiae but also the diseases they cause. Such collaboration is a goal of the European Group on Rickettsiae, Ehrlichiae and Coxiella that was established as a part of the European Society of Clinical Microbiology at its IVth Congress in Nice in 1989. The members of this group met during the IVth International Symposium on Rickettsiae and Rickettsial Diseases.

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