occurring in 1984). However, plague has made an astonishing comeback in the last decade.

Methods: n/a.

Results: After a silence of 50 years, an outbreak of bubonic plague suddenly occurred close to Oran in Algeria. in June 2003. Eighteen bubonic cases were identified, and Yersinia pestis was isolated from 6 patients. In July 2008, a new cluster was reported among nomads 300 km south of the first one. Four members of one family were affected and one died. The bacillus was isolated from one patient. No epidemiological association was identified between the two events. On June 2009, 25 years after the last occurrence in the country, Libya reported five confirmed cases of bubonic plague in the Tobruk area. Y. pestis was isolated from three patients. In all these cases, further local ecological investigations confirmed the existence of a natural focus The re-emergence of human plague in the region is not without international consequences. Two of the last concerned natural foci are close to an international port which raises the question of the potential exportation of infected rodents. Cross-border tensions, between "plague countries" and "plague-free countries" have been observed although the foci's limits are unknown as any systematic ecological investigation and surveillance is lacking. Additionally, the potential weaponization of Y. pestis together with international political tensions feed a recurrent interest in plague in North Africa. False rumors of alleged military laboratory accidents or terrorist acts are routinely mentioned, although events could be first explained by the natural history of the disease.

*Conclusion:* In this context, and although the number of human cases has been very limited so far, the first priorities are to establish appropriate ecological surveillance and agree on a common plague control strategy for the region.

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76.011

Analysis and characterization of mouse monoclonal antibodies reactive to Chikungunya virus (CHIKV)

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*Background:* Chikungunya virus (CHIKV), an alphavirus belonging to the Togaviridae family, is transmitted to human by several species of mosquitoes, with *Aedes Aegypti* and *A. Albopictus* being the two main vectors. The virus is endemic in Africa, India, South-East Asia and recently in southern-Europe and is responsible for an acute infection of abrupt onset characterized by high fever, asthenia, headache, rash, myalgia and a painful polyarthralgia. Because the re-emergence of CHIKV fever, the development of anti-CHIKV monoclonal antibodies (MAbs) is critical for diagnosis and basic research on CHIKV infection. In this

study we analyzed the reactivity of a panel of MAbs to CHIKV.

Methods: A panel of 10 MAbs has been tested for reactivity to CHIKV by indirect ELISA, for which a Maxisorp plate has been coated with purified CHIKV virions. All the MAbs that tested positive in ELISA, were further tested by indirect immunofluorescence assay (EUROIMMUN, Medizinische Labordiagnostika AG). Immunoblot analyses were carried out in order to identify the protein target towards the reactive Mabs are raised to. Furthermore, transient expression of E2 in COS7 cells, followed by fluorescence imaging, has been used for a better identification of MAbs reactive to E2 protein.

*Results*: Among 10 MAbs tested, 5 showed reactivity with CHIKV antigens in indirect ELISA; only 2 of them were further confirmed as reactive by immunofluorescence assay. Immunoblotting analyses, performed with purified CHIK virions, showed that these 2 MAbs had significant reactivity with envelope glycoprotein E1 or E2. Immunofluorscence studies performed on COS7 cells transiently expressing E2 protein, showed that both the anti-CHIKV Mabs targeted specifically viral glycoprotein E2.

*Conclusion:* In this study a panel of 10 Mabs were tested for their reactivity with CHIKV by indirect ELISA, Immunoblot and immunofluorescence assay on cells expressing E2 protein. 2 Mabs specifically targeted to viral glycoprotein E2 were identified. The 2 Mabs might be suitable for developing diagnostic tests other than be helpful for studying the biology of CHIKV and pathogenesis disease. For these 2 Mabs, the potential to neutralize CHIKV infection is under evaluation.

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#### 76.012

Study of Hanta-viral hemorrhagic fever with renal syndrome (HFRS) in an endemic region of Bulgaria

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*Background*: HFRS started registration in Bulgaria since 1953. The major epidemics were first described during 1957 and 1959 in Western Rhodopa region. In the 1953-1991 period 412 patients were registered and 67 of them with lethal exit. The aim of the study was to describe the clinical and epidemiological characteristics of HFRS, and to discuss the professional implications of the disease, as well as its control and prevention.

*Methods*: The authors analyze clinical, laboratory and epidemiological data about 21 cases of HFRS for the period 1987-2009. The cases were etiologically proven in the National Reference Arboviral laboratory. Hantaan (37.5%) and Puumala (12.5%) viruses were identified.

*Results*: All cases were registered in typical season - early spring, summer and late autumn. Males (80.95%) present risk gender group for HFRS related to profession — military servants, construction workers in the endemic mountain regions etc. Mountaineer personnel are exposed to infection when working, digging, eating or sleeping in fields infested by infected rodents.

Patient age varies from 14 to 83  $(38.52 \pm 4.49)$  years and 57.14% of them were between 15–35 years old.