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PROCEEDINGS

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O21: Development of antibodies against Grapevine Pinot gris virus (GPGV) in rabbits and Camelids.

Vianney Poignavent¹, Magadalène Kosfitskas¹, Léa Gerber¹, Christophe Ritzenthaler¹, Serge Muyldermans², Denise Altenbach³, and Christophe Debonneville^{3*}

1 CNRS, Institut de Biologie Moléculaire des Plantes, 12 Rue du Général Zimmer, 67000 Strasbourg, France. 2 Vrije Universiteit Brussel, Department of Bio-engineering Sciences, Pleinlaan 2, 1050 Brussels, Belgium 3 BIOREBA AG, Christoph Merian-Ring 7, CH-4153 Reinach, Switzerland. *Corresponding author: debonneville@bioreba.ch

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After several common grapevine viruses were ruled out (Ampelo-, Nepo-, Clostero-, and Vitiviruses), the study of Pinot gris vines showing symptoms of leaf deformation, stunting, and chlorotic mottling by deep sequencing revealed the presence of a new trichovirus named Grapevine Pinot gris virus (GPGV, Giampetruzzi et al, 2012). The effects of GPGV infection on grapevines are still unclear and the link between virus infection and the occurrence of symptoms is still poorly characterized. Today GPGV has been confirmed to infect at least 28 grape varieties and has been reported in many countries in Europe and Asia as well as in Australia, Canada and USA. So far PCR is the only method available to confirm GPGV infection. A serology-based diagnostic tool is needed to perform cost-effective and robust large scale testing.

A purified recombinant GPGV coat protein produced in E. coli was used to immunize rabbits as well as alpacas (Camelidae) to maximize chances to obtain useful detection reagents. In rabbits classical polyclonal antibodies were then purified to be used as DAS-ELISA reagents. In alpacas small antibody-like structures composed of heavy chain only (VHH, about 100 residues and 15kDa) were screened to isolate those binding to GPGV.

So far classical IgGs purified from immunized rabbits were used in a prototype DAS-ELISA kit. The results obtained using infected grapevine material (GPGV positive; confirmed by PCR) are very promising. The reagents allowed the detection of different GPGV isolates from leaves and wood samples. In addition IgG-like small antibodies obtained from immunized alpacas were identified and are currently being characterized. These VHHonly antibodies have the potential to greatly improve the sensitivity and detection spectrum of the current test. To our knowledge, this is the first DAS-ELISA for the detection of GPGV. Due to the worldwide distribution of the virus, this new reagent will be of great interest for certification programs and diagnostic laboratories.

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Giampetruzzi, A., Roumi, V., Roberto, R., Malossini, U., Yoshikawa, N., La Notte, P., Terlizzi, F., Credi, R., and Saldarelli, P. 2012. A new grapevine virus discovered by deep sequencing of virus- and viroid-derived small RNAs in cv Pinot gris. Virus Res. 163:262-268