## Rodenticide resistance and environmental monitoring

## Twenty years of rodenticide resistance monitoring in Belgium

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Our research into anticoagulant rodenticides (AR) resistance in Flemish rat populations began in 2003 with blood clotting response (BCR) tests, in which small doses of ARs were administered to measure the prothrombin time and assess blood clotting effects. Over three years (2003-2005), we screened 691 rats from various locations for susceptibility to warfarin, bromadiolone, and difenacoum, identifying around 20% of the population as anticoagulant-resistant. Concurrent advances led to the discovery of the Vkorc1 gene and its role in rodenticide resistance. Genetic analysis of 119 rats revealed two prevalent mutations: Y139F and L120Q, each showing unique geographical distributions. Y139F was common in the western and eastern regions of Flanders, while L120Q appeared predominantly in the western central area, with notable absence of resistance in central Flanders. From 2006 to 2010, a second screening was conducted on 845 rats, of which 286 were at least warfarin-resistant. Depending on their BCR test result a total of 449 rats were genetically analysed, confirming the geographic spread of Y139F and L120Q and identifying for Flanders a rare Y139C mutation. Combined BCR and genetic findings highlighted Y139F's association with warfarin and bromadiolone resistance, while L120Q primarily caused warfarin resistance; both mutations showed very limited difenacoum resistance. In 2013, our screening methodology transitioned to a standardized genetic approach, focusing on SNP detection to replace BCR tests due to animal welfare and efficiency concerns. We aimed for a broad geographic spread by limiting samples to one per location due to spatial correlation, and targeted 1200 samples annually. This approach continued from 2013 to 2019, though 2017 and 2018 featured reduced sampling with 500 samples in central Flanders, where resistance prevalence was lower. By the end of this period, overall resistance had risen to 40%, influenced by gradual expansion of the resistant mutations' geographical range and increased local prevalence. Most recently, a 2024 screening revealed preliminary results indicating resistance levels nearing 50%. In some locations, rats were found to carry a combination of Y139F with either L120Q or Y139C mutations. Previous BCR tests in our lab on rats with the Y139F and L120Q combination indicated an increased degree of difenacoum resistance, suggesting potential challenges for rodent control in these areas. To evaluate whether practical implications of resistance have shifted over time, we recently conducted BCR tests on 67 rats from a historically high-resistance location (only with Y139F prevalence), specifically assessing for difenacoum resistance. Results showed that only 5 rats (7.5%) exhibited difenacoum resistance, a proportion that aligns with past findings and remains within acceptable limits for effective rodent control.

In conclusion, resistance has steadily increased over time, now reaching a concerning level of nearly 50%. Nonetheless, effective rodent management is achievable through targeted use of AR compounds based on the specific mutations present. Emphasis on preventive measures, mechanical trapping, and adherence to integrated pest management (IPM) practices will further support sustainable control efforts.

