

## Article

# Biocontrol of Biofilm Formation: Jamming Sessile-Associated Rhizobial Communication by Rhodococcal Quorum-Quenching

Yvann Bourigault <sup>1,2</sup>, Sophie Rodrigues <sup>3</sup>, Alexandre Crépin <sup>4</sup>, Andrea Chane <sup>1</sup>, Laure Taupin <sup>3</sup>, Mathilde Bouteiller <sup>1,2</sup>, Charly Dupont <sup>1,2</sup>, Annabelle Merieau <sup>1,2</sup>, Yoan Konto-Ghiorghi <sup>1,2</sup>, Amine M. Boukerb <sup>1</sup>, Marie Turner <sup>5,6</sup>, Céline Hamon <sup>5</sup>, Alain Dufour <sup>3</sup>, Corinne Barbey <sup>1,2</sup>, and Xavier Latour <sup>1,2,6\*</sup>

<sup>1</sup> Laboratory of Microbiology Signals and Microenvironment (LMSM EA 4312), University of Rouen Normandy, 55 rue Saint-Germain, F-27000 Evreux, France; yvann.bourigault@univ-rouen.fr (Y.B.); corinne.barbey@univ-rouen.fr (C.B.); chane.andrea@gmail.com (A.C.); mathilde.bouteiller7@univ-rouen.fr (M.B.); charly.dupont7@univ-rouen.fr (C.D.); annabelle.merieau@univ-rouen.fr (A.M.); yoan.konto-ghiorghi@univ-rouen.fr (Y.K-G); amine.boukerb@univ-rouen.fr (A.M.B.)

<sup>2</sup> Research Federations NORVEGE Fed4277 & NORSEVE, Normandy University, F-76821 Mont-Saint-Aignan, France

<sup>3</sup> Université de Bretagne-Sud, EA 3884, LBCM, IUEM, F-56100 Lorient, France; sophie.rodrigues@univ-ubs.fr (S.R.); laure.taupin@univ-ubs.fr (L.T); alain.dufour@univ-ubs.fr (A.D.)

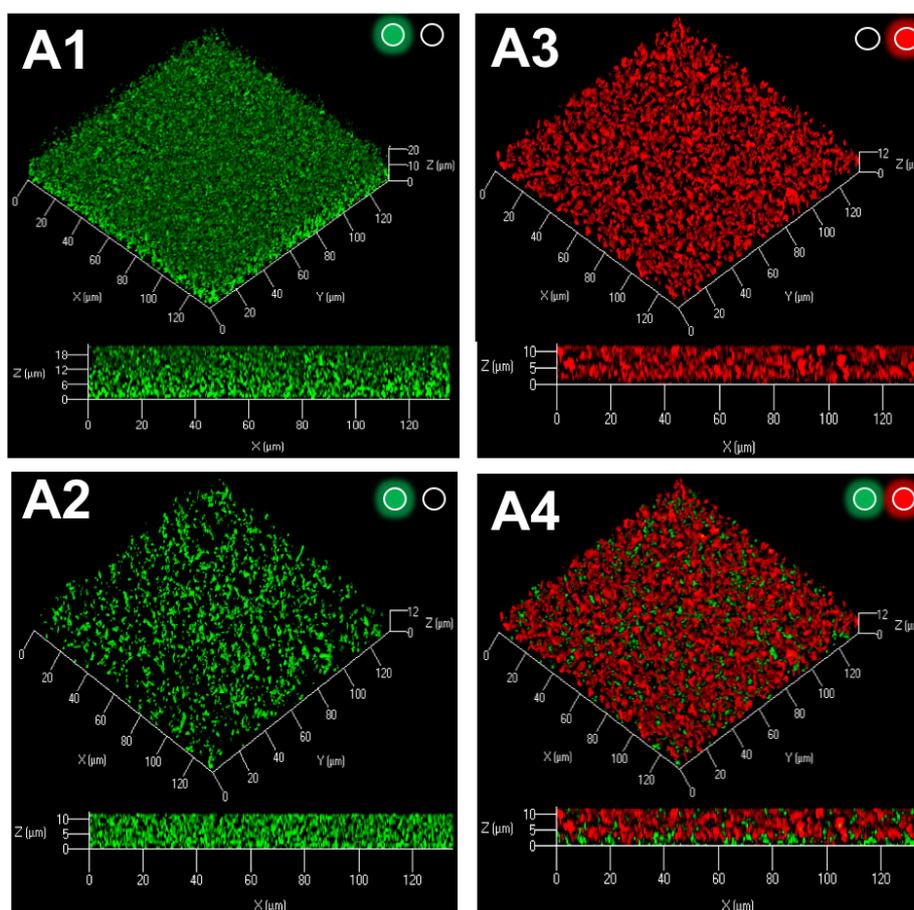
<sup>4</sup> Laboratoire Ecologie et Biologie des Interactions, UMR CNRS 7267 F-86073 Poitiers, France; alexandre.crepin@univ-poitiers.fr (A.CR)

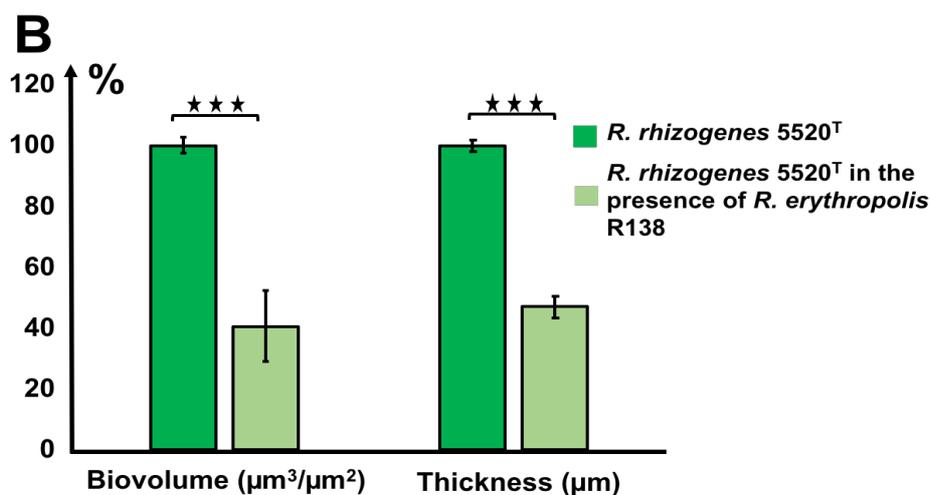
<sup>5</sup> Végénov, F-29250 Saint-Pol-de-Léon, France; turner@vegenov.com (M.T.); hamon@vegenov.com (C.H.)

<sup>6</sup> Biocontrol Consortium, F-75007 Paris, France

\* Correspondence: xavier.latour@univ-rouen.fr; +33-235-146-000 (X.L.)

## Supplementary Material





**Figure S5. Inhibition of rhizobial biofilm formation by *Rhodococcus erythropolis* R138 (ratio 1:10).** Confocal laser scanning microscopy (CLSM) analysis of the *R. rhizogenes* 5520<sup>T</sup> biofilm (A1) or dual-species biofilm formed by *R. rhizogenes* 5520<sup>T</sup> and *R. erythropolis* R138 (A2,A3,A4) was achieved at an inoculation ratio of 1:10. *R. rhizogenes* and rhodococcal bacteria were tagged with GFP and mCherry via the pHC60-*gfp* and pEPR1-*mCherry* vectors, respectively. 3D shadow representation and side view of the biofilm produced by *R. rhizogenes* alone (A1), and by a mixed culture of *R. rhizogenes* and *R. erythropolis* (A2) analyzed in the green channel. Location of rhodococcal cells is revealed by 3D shadow representation and side view of the biofilm produced by the mixed culture analyzed in the red (A3) or red plus green channels (A4); (activation of the green or red channel is indicated by a light spot of the corresponding color). COMSTAT analyses of *R. rhizogenes* green fluorescence in single and dual-species biofilms. *R. rhizogenes* biofilm biomass and thickness values were normalized (set to 100%) as a reference for a comparison with mixed biofilm conditions (B). The data shown are the means of three measurements from three independent experiments. Significant differences (Mann and Whitney test;  $p$ -value < 0.01) in biovolume and thickness are indicated by asterisks (\*\*\*  $P$  < 0.001).