



FINAL PUBLISHABLE SUMMARY REPORT

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1. Final Publishable Summary Report



R-GNOSIS

**Resistance in Gram-Negative Organisms:
Studying Intervention Strategies**

1.1 Executive Summary

Antibiotics are a mainstay of public health, but their use has increased exponentially leading to the emergence of antibiotic resistance. The European community and health care settings are facing a dramatic increase in infections caused by MDR-GNB, with few effective therapeutic options remaining in the armamentarium of clinicians. There is a striking lack of new antimicrobial agents against MDR-GNB. Even if new agents were to be discovered, the lead in time is considerable as there are no truly new agents expected on the market in the short or medium term. Even if new agents were to come to market, on their own, they could not solve the resistance problem. Effective measures to contain resistance and limit the spread of MDR-GNB are therefore urgently needed to protect the health and well-being of the people of Europe and world-wide.

R-GNOSIS – “Resistance in Gram-Negative Organisms: Studying Intervention Strategies” is a European collaborative research project awarded 11.9 Million Euro funding from the European Commission under the Seventh Framework Programme (FP7) for Research and technology.

Lasting 5,5 years, R-GNOSIS aimed at determining the efficacy and effectiveness of cutting-edge interventions to reduce carriage, infection and spread of Multi-Drug resistant Gram-negative Bacteria (MDR-GNB).

The project combined five international clinical studies, all supported by highly innovative microbiology, mathematical modelling and data management.

The five clinical studies investigated the following interventions:

- A Point-Of-Care-Testing guided management strategy to improve appropriate antibiotic prescription for uncomplicated UTI in primary care
- Gut decolonization followed by fecal microbiota transplantation in outpatients with intestinal carriage of MDR-GNB
- A “test and prescribe” strategy, based on rapid diagnostic testing of faeces for MDR-GNB to optimize antibiotic prophylaxis in colo-rectal surgery
- Contact Isolation of patients with ESBL-producing Enterobacteriaceae in general hospital wards
- Three Decolonization strategies in ICUs

Seven laboratories across Europe performed microbiological analyses, as well as unique quantitative experiments. All information were integrated by three groups of mathematical modellers into highly innovative models to better understand and predict future trends and effects of interventions.

The studies and analyses proposed in R-GNOSIS will generate a step-change in identifying evidence-based preventive measures and clinical guidance for primary care and hospital-based physicians and health-care authorities, to combat the spread and impact of infections caused by MDR-GNB in Europe.

R-GNOSIS was coordinated by Marc J. Bonten (UMC Utrecht) and brought together a multidisciplinary team with complementary expertise of clinicians, microbiologists and epidemiologists, infectious disease specialists, caregivers in inpatient care and mathematical modellers. The R-GNOSIS consortium was made of 24 partners from 9 countries which include United Kingdom, Denmark, Germany, Netherlands, Belgium, France, Spain, Switzerland and Israel.

1.2 Project context and objectives

The European community and health care settings are facing a dramatic increase in infections caused by MDR-GNB, with few effective therapeutic options remaining in the armamentarium of clinicians. There is a striking lack of new antimicrobial agents against MDR-GNB. Even if new agents were to be discovered, the lead in time is considerable as there are no truly new agents expected on the market in the short or medium term. Even if new agents were to come to market, on their own, they could not solve the resistance problem. Effective measures to contain resistance and limit the spread of MDR-GNB are therefore urgently needed to protect the health and well-being of the people of Europe and world-wide. We are confronted with a formidable enemy, equipped with sophisticated molecular methods to express and exchange resistance genes, capable of colonizing multiple reservoirs, and harbouring a bewildering array of virulence factors to infect any suitable host, both the hospitalised frail and healthy people living in the community.

The R-GNOSIS project aimed at identifying the most effective measures for controlling selection and transmission of MDR-GNB and their genes, the most effective measures to reduce infections caused by MDR-GNB in relevant patient populations in the community and healthcare settings, optimizing treatment and prophylaxis strategies to avoid the detrimental consequences of these bacteria on patient outcome and investigating the critical molecular aspects for persistence and transfer of resistance genes in the human gut.

The scope of the problem (an estimated 25,000 people dying from infections caused by multiresistant bacteria in Europe in 2007, with infections caused by MDR-GNB increasing exponentially) means that ‘more of the same’ research is simply not an option. An “out-of-the-box” conceptual step-change is necessary, in which the evidence base is optimised for “well known but inadequately researched” infection prevention strategies through innovative and state-of-the-art study designs, and in which counterintuitive and highly innovative, solutions are identified and rigorously evaluated.

In this perspective, **the main objective of the R-GNOSIS (Resistance in Gram-Negative Organisms: Studying Intervention Strategies) project was to determine – in the most relevant patient populations – the efficacy and effectiveness of cutting-edge interventions to reduce acquisition, carriage, infection and spread of Multi-Drug Resistant Gram-negative Bacteria (MDR-GNB).**

To meet this objective, the project combined five international clinical intervention studies – all supported by highly innovative microbiology and mathematical modelling.

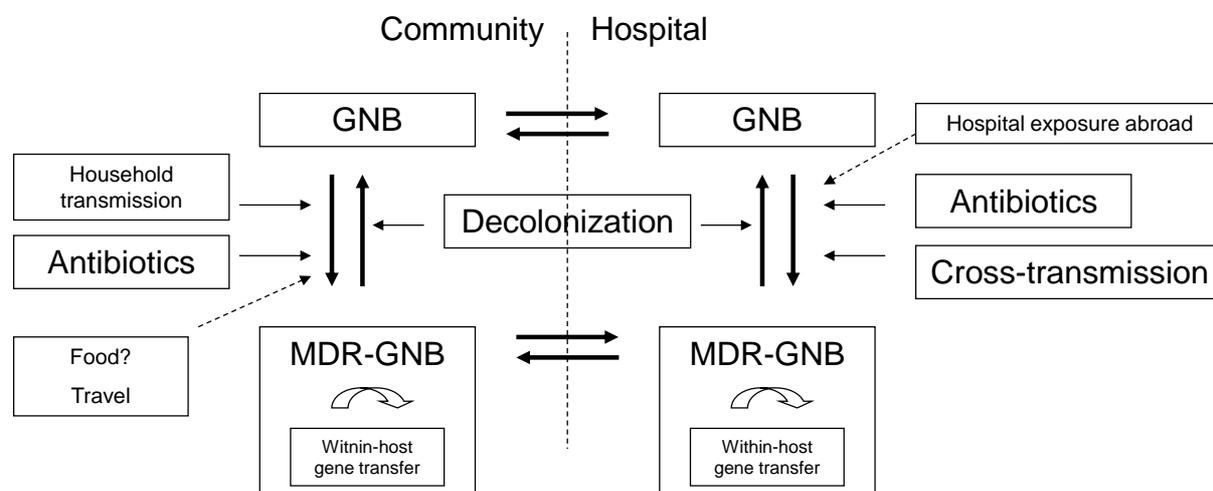


Figure 1: Transmission of MDR-GNB or their resistance genes

The five clinical studies investigated the following interventions, with the following specific objectives:

- 1) A Point-Of-Care-Testing guided management strategy to improve appropriate antibiotic prescription for uncomplicated UTI in primary care (WP2)
 - To quantify the effects of an innovative POCT guided treatment approach on reducing inappropriate antibiotic use in women with UTI in a RCT in primary care
- 2) Gut decolonization followed by fecal microbiota transplantation in outpatients with intestinal carriage of MDR-GNB (WP3)
 - To quantify the effects of a highly innovative decontamination/recolonisation approach in intestinal carriers of MDR-GNB on intestinal carriage with MDR-GNB in a randomized trial
- 3) A “test and prescribe” strategy, based on rapid diagnostic testing of faeces for MDR-GNB to optimize antibiotic prophylaxis in colo-rectal surgery (WP4)
 - To quantify the effects of a “rapid screen and modify surgical prophylaxis” strategy on the exposure of patients undergoing colo-rectal surgery to inappropriate antibiotics and the occurrence of SSIs caused by MDR-GNB in an interrupted longitudinal study
- 4) Contact Isolation of patients with ESBL-producing Enterobacteriaceae in general hospital wards (WP5)
 - To quantify the incremental effects of isolation of ESBL-E-carriers to an improved hand hygiene program alone on ESBL-E infection rates in general hospital wards in a cRCT
- 5) Three Decolonization strategies in ICUs (WP6)
 - To quantify the incremental effects of three different decolonization strategies to standard care procedures (including chlorhexidine body washings) on infection and transmission rates of MRDR-GNB in ICUs in a cRCT

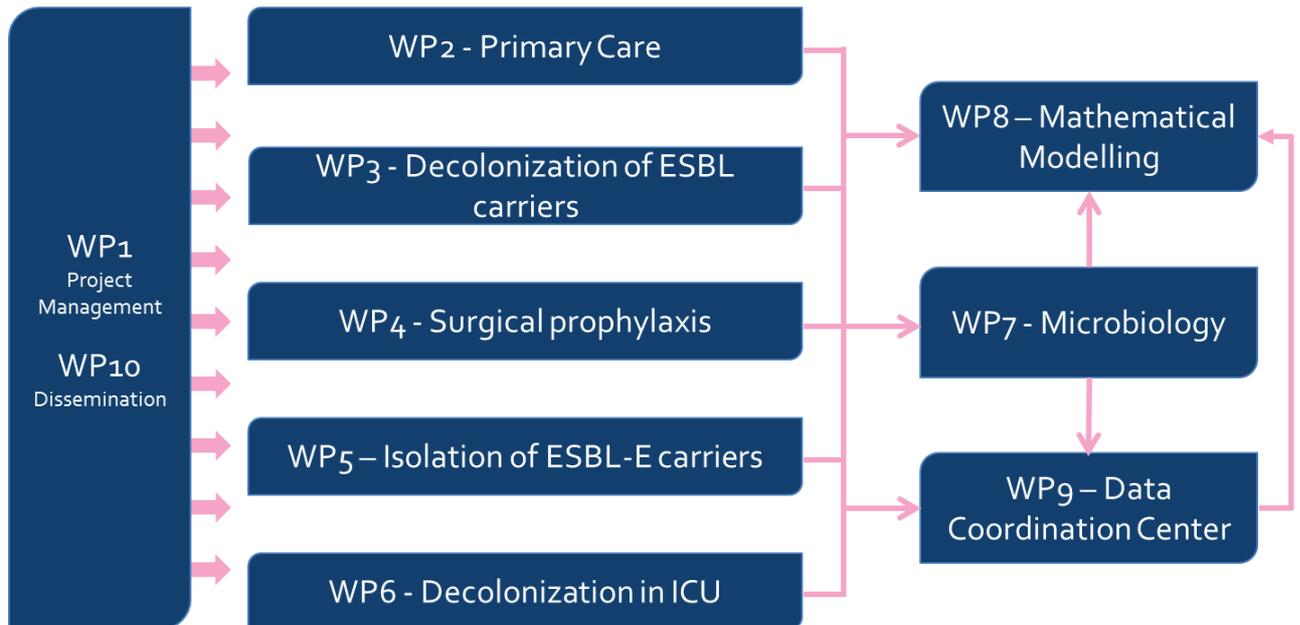
These studies were supported by the microbiology WP (WP7) aiming at identifying emerging MDR-GNB clones and resistance genes in the clinical trials and investigating the underlying microbiological mechanisms for successful spread; and by the mathematical modelling WP (WP8) aiming at developing and updating multi-compartment models to create applied modelling tools for predicting (long-term) effects of interventions, using data from the clinical trials and at investigating the dynamics of MDR-GNB using detailed microbiological data of intestinal microbial interaction, derived from the clinical trials.

Embedded within these clinical trials R-GNOSIS executed observational and case-control studies to determine associations between intestinal and respiratory tract carriage with MDR-GNB and infection.

All clinical studies will progress science beyond the state-of-the-art in generating new and translational clinically relevant knowledge, through hypothesis-driven studies with a focus on patient-centred outcomes that matter to the people of Europe and beyond.

The studies and analyses proposed in R-GNOSIS will generate a step-change in identifying evidence-based preventive measures and clinical guidance for primary care and hospital-based physicians, as well as health-care authorities, to combat the spread and impact of the unprecedented rise of infections caused by MDR-GNB in Europe.

The overall structure and interactions of the WPs are demonstrated in the following figure. WP1 and WP10 are support packages and interact with all other WPs:



1.3 The main S&T results/foregrounds

1.3.1 WP2 – POETIC: Point of care testing for urinary tract infections in primary care



Serious gram negative infections are becoming increasingly common, and the prevalence of antibiotic resistant pathogens is increasing, making these infections harder to treat, with increasing patient morbidity and mortality. Unnecessary antibiotic prescribing in primary care is one probable factor in driving the increasingly antibiotic resistant gram negative infections. At the present time, the presentation (including the microbiology and pathogen sensitivities), management (including empiric antibiotic prescribing), and outcomes of uncomplicated urinary tract infection (UTI) in primary care in Europe is not well described. Most women presenting with uncomplicated UTI are prescribed antibiotics despite evidence that some do not benefit from antibiotics and probably have non-infectious explanations for their symptoms. A rigorous description of current practice would help identify targets for improved clinical practice.

Point of care tests (POCT) are often introduced into routine care before clinical and cost effectiveness in terms of impact on patient outcomes has been demonstrated. Point of care urine culture is already in common use in everyday general practice in some European settings, for example Denmark, but have never been subjected to rigorous randomised controlled trials. The opportunities and barriers to the uptake of point of care tests in primary care for UTI in contrasting European settings have not been well described. This work package therefore undertook a four-phase program of study to address the following questions:

- Stage 1: an initial pilot study, conducted in Wales only, to assess and further develop study procedures, including the use of the POCT, in preparation for an RCT.
- Stage 2: an observational stage where clinicians recruited sequential patients with UTI symptoms, recording their usual-care diagnostic procedures, treatment and sampling.
- Stage 3: a randomised controlled trial (RCT), where patients were randomised to either the POCT guided management strategy (Flexicult™) or to a standard UTI management strategy based on best local guidelines.
- Stage 4: a qualitative study which explored the barriers and opportunities for use of POCTs with clinicians.

The overall aim of the POETIC study was **to describe the presentation, management and clinical outcomes of women presenting with uncomplicated UTI, and to evaluate the management of suspected uncomplicated UTI in women presenting in primary care through the use of a novel Point of Care Test (POCT) guided urinary tract infection (UTI) management strategy (Flexicult™)**. We also explored barriers and opportunities of using a POCT for the management of UTI in primary care with clinicians.

The POCT provides clinicians, at the point of care, within 24 hours, with a diagnosis of bacterial UTI (or not) and resistance profiles of any identified pathogen to the antibiotics most commonly used for UTI in primary care. The study aimed to determine whether or not this information aided clinicians to more appropriately prescribe antibiotics for uncomplicated UTI's (i.e. minimise the use of antibiotics for women where no bacterial infection is identified, and ensure the narrowest spectrum antibiotic appropriate to the sensitivity of the infecting organisms was prescribed when a bacterial infection is identified). Appropriateness was assessed against the result of laboratory based analysis of the urine sample collected at presentation.

The study was conducted in four European primary care research networks (England, Wales, the Netherlands and Spain).

The main findings of these studies were:

- Stage 1: We developed a feasible intervention for use in four European settings, and a plan for evaluation.
- Stage 2: We found high levels of variation in antibiotic prescribing despite only small differences in microbiology, presentation and populations.
- Stage 3: Use of the Flexicult™ POCT test was neither clinically nor cost effective as an addition to the primary care management of UTI when used primarily to confirm or modify initial empirical antibiotic prescribing decisions.
- Stage 4: The relative speed of obtaining confirmation of UTI and indication of appropriate antibiotics to prescribe, was seen as a key advantage of the Flexicult™ POCT. However, staff requirements and timing of use of the test were perceived as important barriers to use.

We have taken the field forward by identifying targets for interventions to improve practice, including the need for better targeted antibiotic treatment and opportunities for symptomatic treatment.

Flexicult™ POCT used in the way used in our trial it was not cost effective, thus our data will re-direct its use in primary care to ensure that POCTs should not be introduced unless their place in clinical pathways is well described and cost effectiveness demonstrated. Critically, we need to determine the cost effectiveness of point of care culture for guiding initial empirical antibiotic prescribing.

Future microbiological research should focus on what causes symptoms in those who have urine that is culture negative. Are there non culturable organisms that cause these symptoms, or do organisms die before they are inoculated onto culture medium?

1.3.2 WP3 – Decolonization/recolonisation in carriers of ESBL- and carbapenemase producing Enterobacteriaceae

Initially, WP3 foresaw a randomized, placebo-controlled, double-blind phase II trial to evaluate the safety and efficacy of colistin and neomycin delivered directly to the colon via a newly developed drug delivery system developed by a French biotechnology company – Da Volterra – (DAV-148) with subsequent application of a probiotic (*Escherichia coli* Nissle 1917) for decolonization of adult carriers of extended spectrum beta-lactamase producing Enterobacteriaceae (ESBL-E). The drug delivery system for DAV-148 was designed to release colistin and neomycin directly in the colon, thereby achieving high concentrations of these antibiotics at the site of ESBL-E colonization and potentially also avoiding the reported side effects in the upper gastrointestinal tract gut. The planned treatment strategy included a “restoration treatment” by a probiotic *E. coli* strain after the conclusion of the intestinal antimicrobial treatment.

Unfortunately, in September 2012, due to a genotoxic potential of this Nissle strain and for some other reasons, Da Volterra decided to stop the further development of DAV-148. This decision made the conduct of the clinical trial in the initially proposed form impossible and after discussion an alternative academic, investigator-initiated trial was designed, using “conventional” colistin sulfate instead of DAV-148 as decolonization regimen and **fecal microbiota transplantation (FMT)** instead of *E. coli* Nissle 1917 as recolonization regimen.

FMT consists in the gastrointestinal application of stool from healthy donors (most commonly family members) via colonoscopy, nasogastric tube or enema with the aim to restore the protective microbiome of natural colonic flora. Cure rates of over 90% have been reported for recurrent *Clostridium difficile* infection (CDI).

The primary objective of R-GNOSIS WP3 was to test the efficacy and safety of colistin sulfate / neomycin sulfate followed by fecal microbiota transplantation (FMT) to eliminate the carriage of ESBL- and / or carbapenemase producing Enterobacteriaceae in 64 adult patients.

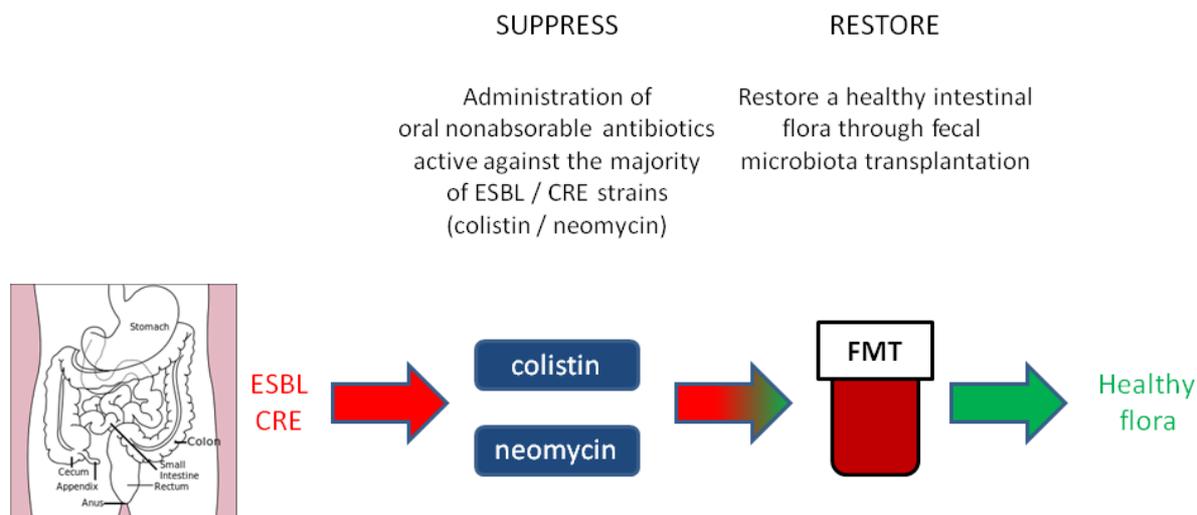


Figure 2: FMT actions

To our knowledge, R-GNOSIS WP3 is the first randomized trial examining the use of FMT for decolonization of multidrug-resistant organisms (MDRO).

However, this complex trial proved to be very challenging and faced multiple challenges and obstacles (regulatory problems, difficulties in the provision of study antibiotics, difficulties with patient recruitment, etc.) that lead to a significant delay in the start of the trial.

Between February 2016 and June 2017, 39 patients have been randomized (16 in Paris, 14 in Geneva, 7 in Utrecht and 2 in Tel Aviv). The last follow-up for the last patient will take place in December 2017 and the analysis of the trial will occur in the first trimester of 2018. The metagenomic analysis of a selected number of samples is ongoing.

1.3.3 WP4 – Surgical prophylaxis

Surgical site infections (SSI) are a common complication after colorectal surgery, associated with increases in morbidity, mortality, length of hospital stay and healthcare costs. SSI rates between 10 and 25% are most commonly reported in prospective clinical trials. These infections can be dramatically reduced by adequate perioperative antibiotic prophylaxis.

The WP4 trial was a multi-centre clinical controlled trial, designed as a before-after study with nested case-control studies with the primary objective of **testing the hypothesis that identification of carriers of ESBL-PE, followed by targeted adaptation of pre-surgical prophylaxis for ESBL-PE carriers, reduces SSI among carriers with minimal harmful effects.**

During the first phase (standard care), ESBL-carriage status was determined, but results were not available at time of prophylaxis. All patients received routine prophylaxis with cefuroxime/metronidazole (I.V. cefuroxime 1.5 gram + metronidazole 500 mg 30 minutes before surgery). ESBL-carriage status was available only after surgery.

During the second phase (intervention), ESBL carriage status was determined and ESBL-carriers received ertapenem prophylaxis (I.V. ertapenem 1 gram, 30 minutes before surgery).

Three groups were studied in nested case-control studies:

- ESBL carriers in Phase 1 who received cefuroxime/metronidazole prophylaxis (ESBL control group)
- ESBL carriers who received ertapenem prophylaxis (ESBL intervention group)
- Non-ESBL carriers who received cefuroxime/metronidazole prophylaxis (non-ESBL control group).

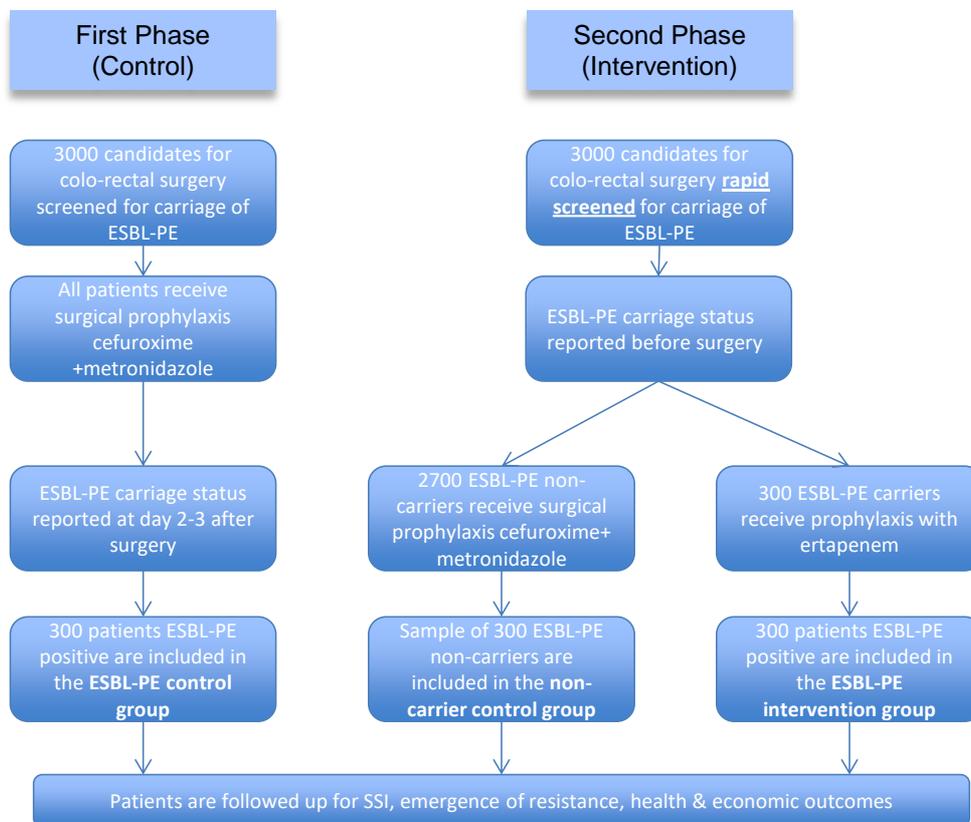


Figure 3: Surgical prophylaxis study scheme

As of the end of March 2017, 3.593 patients have been recruited in the 3 study sites (Tel Aviv – TASMC, Geneva – UNIGE, Belgrade – CCS). They were screened for ESBL carriage before undergoing colorectal surgery using standardized protocol. The percentage of ESBL positive patients calculated from the first screening performed is 27.1% in TASMC, 11.3% in UNIGE and 8.7% in CCS. Risk factors for ESBL carriage among the target population have been described and it was determined that the risk of SSI among ESBL carriers who receive routine antibiotic prophylaxis (cephalosporin + Metronidazole) is much higher than among non-carriers 23% vs 11%. The feasibility of the "screen and treat strategy" was shown, even without the availability of rapid test, using pre-surgical appointment as the opportunity to screen with less than 10% misses. Also it was shown that the "screen and treat strategy" results in dramatic reduction in SSI rates among ESBL carriers (from 23% to 12.8%); with similar effect on deep/organ space SSI. Furthermore, the WP4 study was able to show that ESBL HAI and SSI occur almost exclusively in ESBL carriers, and the risk among carriers who receive routine prophylaxis is 10 fold higher than among non-carriers. Parallel reduction in ESBL SSI by "screen and treat strategy" was also shown.

1.3.4 WP5 – Patient isolation strategies for ESBL-E carriers in medical and surgical hospital wards

“To isolate or not to isolate” patients with ESBL-producing Enterobacteriaceae is currently one of the most controversial questions in the field of infection control (IC). Most national guidelines recommend contact isolation (CI) of patients with MRSA (Methicillin-resistant Staphylococcus aureus) or VRE (Vancomycin-resistant Enterococci). For ESBL, the evidence is less conclusive and is not investigated in wards with endemic ESBL levels but instead is often only derived from outbreak investigations. A recently published systematic review to examine the efficacy of infection control interventions for the control of ESBL in hospital in non-outbreak settings identified only four uncontrolled retrospective studies.

The importance of the environment as an intermediary in the transmission of multidrug resistant organisms (MDRO) is also poorly understood. In general, Enterobacteriaceae lose viability quickly and are recovered less frequently from hospital environments than Gram-positive organisms. This characteristic does not support isolation in single rooms. However, a recent study found substantial contamination of gloves and gowns with multidrug-resistant bacteria, thus supporting the use of gloves and gowns for treating these patients.

Standard precautions (SP) may be as effective for limiting the spread of ESBL, especially when hand hygiene compliance is high, and one study even found lower hand hygiene compliance under isolation conditions.

In addition, high ESBL admission rates make it virtually impossible to focus isolation on patients identified as ESBL carriers only by microbiological samples obtained for clinical purposes because this identification method leaves many ESBL-carriers undetected.

Hence, a combined approach of active surveillance screening at admission and contact isolation of detected ESBL-carriers would be the logical consequence. However, this would increase the management costs for ESBL patients even further.

Implementing HICPAC guideline recommendations would have considerable cost implications and impact on the quality of patient care. Therefore, adding CI measures for the rapidly increasing number of patients colonized with ESBL should be more evidence based.

In this perspective, the aim of the WP5 study was to **determine the benefits of contact isolation (CI) over standard precautions (SP) for the care of ESBL-E-carriers in non-ICUs**. This cluster-randomized controlled clinical trial with a cross-over design was carried out in 20 non-ICUs in 4 university hospitals in Berlin (Germany), Geneva (Switzerland), Madrid (Spain) and Utrecht (Netherlands) between January 2014 and August 2016. All wards were required to apply both infection control strategies (CI and SP) for an entire year each in a randomized order. Thus, they started with the first infection control strategy in the first and switched to the second infection control strategy in the second year of participation. In order to ensure reliable study results, strict adherence to the study protocol was necessary and had to be monitored closely during the entire course of the study.

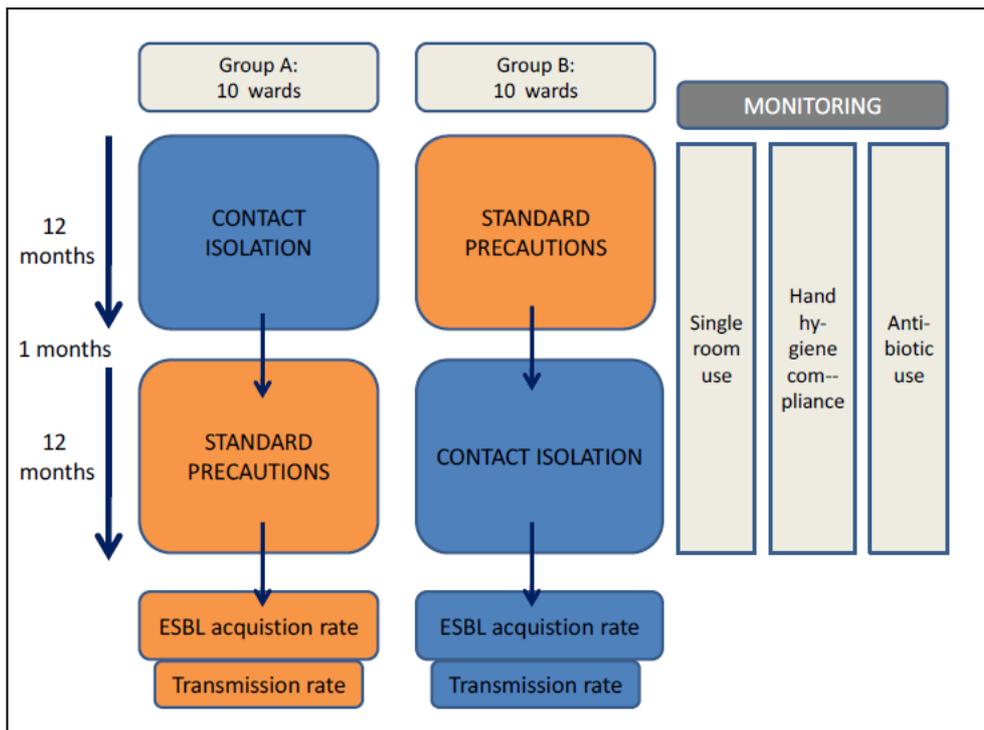


Figure 4: WPS Study design

Final conclusions are not yet possible, as the databases are still open and adjusted analyses have not been carried out. ESBL-E admission prevalence and incidence are much higher than previously thought. Considering the raw and unadjusted data, CI does not significantly reduce the incidence density of ESBL-E-acquisition among hospitalized patients in adult non-ICU wards in hospitals with universal ESBL-E admission and discharge screening. For ESBL-producing *Klebsiella pneumoniae* there was a tendency in favour of CI over SP, while the incidence of ESBL-producing *Escherichia coli* was even higher in CI compared with SP. However, the differences were not significant.

ESBL-E-acquisitions may be caused by patient-to-patient transmissions, but may also reflect selective shifts in the intestinal microbiota during e.g. an antimicrobial treatment. First molecular PCR analyses of ESBL-genes in a small subset of possible transmission events reveal that approximately 70% of ward-acquisitions were probably not caused by patient-to-patient transmission. Whole genome sequencing bring further insight on transmission events between patients in a larger dataset.

Publications of the main study results are expected in the course of the next year.

1.3.5 WP6 – Decolonization strategies in Intensive Care

Successful suppression of ESBL carriage and other MDR-GNB could be an effective infection control measure, as it may not only protect the individual patient by reducing the risk of infection, but also the population at large by reducing transmission of MDR-GNB. There is conflicting evidence on the effectiveness and safety of such an approach in intensive care units (ICU).

In R-GNOSIS, WP6 investigated this approach. The aim of the WP6 cluster-randomised study was to **investigate the effectiveness and safety of three decontamination strategies for elimination of MDR-GNB in patients in the intensive care unit.**

Although decontamination strategies with antibiotics, such as selective digestive tract decontamination (SDD) and selective oropharyngeal decontamination (SOD) have been used in intensive care patients since 1984 in settings with relatively low levels of antibiotic resistance, there is conflicting evidence on the effects on patient survival and antibiotic resistance in other settings.

For years, the beneficial effect of SDD and SOD on patient outcomes was only apparent in meta-analyses. More recent studies however, mainly performed in the Netherlands, showed a significant benefit for patients treated with SOD and SDD. However, large trials in Europe are lacking.

Another question is the relative effectiveness of strategies using antiseptics, as opposed to antibiotics, such as chlorhexidine mouthwash. These have never been compared head-to-head. Antiseptics might prove a more effective strategy to eliminate MDR-GNB at a lower risk of developing resistance.

To answer these questions a European cluster randomised trial was designed, with crossover of three decontamination interventions: SDD, SOD and chlorhexidine mouthwash. The effects of these interventions on clinical outcomes such as survival of ICU patients and ecological effects, such as the development of antimicrobial resistance and influence on cross-transmission of MDR-GNB, were compared to a baseline period, in which no decontamination strategy was implemented.

WP6 has also experienced significant delay with the study start due to unexpected hurdles emerging in the process of obtaining ethical approval in several European countries.

In total, 13 hospitals had been recruited in six European countries. During the last 18-month period of the project, patient recruitment continued; the last two sites stopped to enrol patients in May 2017. Each centre participated in the study for 27 months. All patients in the ICU with an expected duration of mechanical ventilation of 24 hours or more were included in the study, increasing the generalizability of the results. Moreover, all patients in the ICU, including those who did not undergo decontamination, took part in monthly point prevalence surveys, for repeated assessment of the prevalence of antibiotic-resistant microorganisms.

Since the start, 9.507 patients have been included. This sample size will increase and will be sufficient to analyse the clinical impact of the decontamination strategies and the ecological effects of each of the interventions. Due to oral mucosal adverse events in 9.8% of 295 patients treated with chlorhexidine 2% mouthwash, the mouthwash was replaced with a chlorhexidine 1% oral gel in the first quarter of 2015.



Figure 5: Study scheme per participating ICUs

The results of the WP6 crossover study will help to guide clinicians to use the most effective decontamination strategy with the lowest risk of antimicrobial resistance in mechanically ventilated ICU patients and help in the understanding of the ecological effect of decontamination strategies in the ICU.

The effectiveness of antibiotic strategies SDD and SOD will also be compared to chlorhexidine mouthwash, an antiseptic strategy which is nowadays widely used. If antiseptic strategies are as effective as strategies using antibiotic prophylaxis, antiseptics would be preferred for their lower risk of developing resistance to antibiotics.

The study will identify the most effective decontamination strategy to fight the spread of resistant MDR-GNB and HRMO, both within and between patients. Results of the final analysis as described in this document are expected in November 2017.

1.3.6 WP7 – Functional microbiology & within-host transmission dynamics of genes, plasmids and clones of MDR-GNB

The overall objective of WP7 was to provide microbiological support to the clinical trials (WPs 2-6) and analyse the quantitative changes and within-host dynamics of MDR-GNB in the presence and absence of antibiotic selection pressure and gut decolonization and to study the emergence, persistence, resistance mechanisms, and clonal relationships of MDR-GNB as pathogens or commensals in the human gut microbiome.

In this perspective, the following investigations and actions were performed during the project duration:

- Supported the development of microbiologic protocols and the writing of laboratory manuals
- Supported the development/ refinement and preclinical validation of novel assays for detection of MDR-GNB that would be utilized as interventions
- Performed preparatory activities for the analysis of samples and strains that will come from the clinical trials
- Assessed a microbiological method to understand the potential role of gastrointestinal colonization with MDR-GNB in patients and the persistence of these organisms
- Analysed the quantitative changes and within-host dynamics of MDR-GNB in the presence and absence of antibiotic selection pressure and gut decolonization
- Studied the emergence, persistence, resistance mechanisms, and clonal relationships of MDR-GNB as pathogens or commensals in the human gut microbiome.

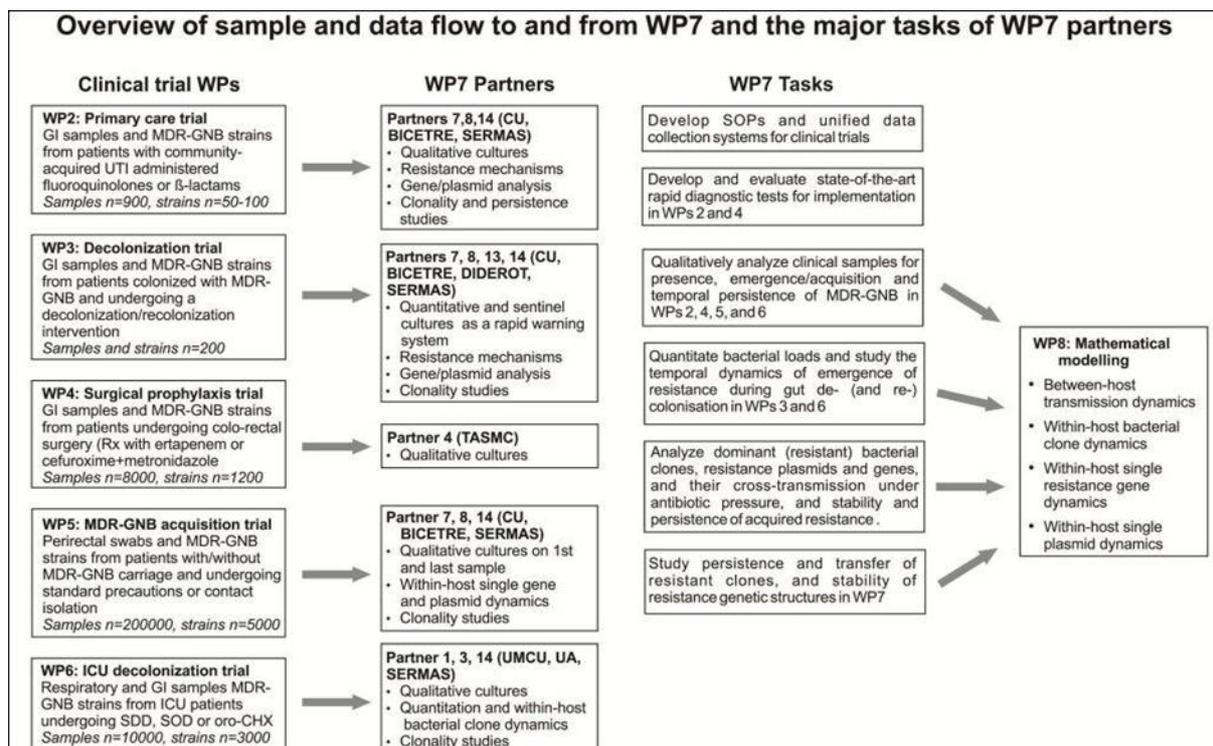


Figure 6: Overview of samples and data flow to and from WP7

1.3.7 WP8 – Mathematical Modelling

The Mathematical Modelling WP worked on the following topics to achieve the primary objective of developing a sound mechanistic and quantitative understanding of the ecological and evolutionary processes determining the within-host dynamics of MDR-GNB and the between-host hospital transmission of these organisms accounting for the resistance gene transfer:

- To study how antibiotics effect the within-host spread of CTX-m resistance genes and to develop to develop mechanistic mathematical models to describe within-host dynamics of MDR-GNB in the human intestine
- To quantify the within-hospital spread of MDR-GNB in hospital settings
- To develop mechanistic mathematical models to describe the between-host transmission dynamics of MDR-GNB in hospitalised patients
- To develop household models for the community transmission and persistence of MDR-GNB
- To evaluate the key factors determining the short- and long-term outcomes of different intervention strategies to control the spread of MDR-GNB, and to quantify the likely long-term outcomes of interventions

1.4 The potential impact & Dissemination of the results

1.4.1 Potential impact of the project

The European Centre for Disease Prevention and Control and the European Medicines Agency estimated that 193,300 patients were infected with MDR-GNB in 2007, and that these infections caused 18,200 excess deaths and 1,375,000 additional hospital days in EU Member States, Iceland and Norway. Moreover, the rate of infections caused by MDR-GNB is still increasing. Today, 'more of the same' research is therefore no longer an option. An "out-of-the-box" conceptual step-change is necessary. Evidence base needs to be optimised for "well known but inadequately researched" infection prevention strategies through innovative and state-of-the-art study designs. Counterintuitive and highly innovative, solutions need to be identified and rigorously evaluated.

The R-GNOSIS programme concentrated on

- the reduction of the prescription of antibiotics by family practitioners;
- preventive administration of antibiotics to IC patients;
- the usefulness of isolating patients who are carriers of ESBL bacteria and
- the prevention of infections following abdominal operations.

R-GNOSIS performed five pivotal international clinical intervention studies, each yielding a clear-cut solution, readily implementable in clinical practice, if proven effective. R-GNOSIS proposes innovative technology and a beyond state-of-the-art hypothesis-driven clinical approach.

Furthermore, R-GNOSIS applied a 'bedside-to-bench' translational approach to study the impact of antibiotics and gut decolonization on MDR-GNB pathogens and commensals in the community or hospital utilizing state-of-the-art microbiological tools. Due to significant unexpected delays in executing highly-innovative clinical trials internationally, the results of two clinical trials were not available within the funding period.

Finally, R-GNOSIS facilitated ground-breaking modelling studies, in which nosocomial transmission capacities for different species of MDR-GNB during different interventions was quantified. Within-host models of bacterial dynamics and gene transfer (based on *in vivo* and *in vitro* experiments) were combined with between-host models, both for hospitals and the community. These integrated models can now be used to provide projections of long-term impacts of interventions.

The R-GNOSIS project will provide a comprehensive knowledge base on the prevention of spread and the management of infections caused by MDR-GNB. With five highly ambitious clinical trials, all supported by state-of-the-art microbiological methods and all interacting with mathematical modelling, R-GNOSIS will deliver guidance for (a) managing infections, (b) selecting appropriate antibiotics for treatment and prophylaxis (c) preventing spread and will also enhance our understanding of the underlying principles of MDR-GNB epidemiology and within-host transfer of resistance genes through integrating modelling and microbiology. The full spectrum of the human health care system is integrated in R-GNOSIS: from reducing inappropriate antibiotic use in primary care to preventing infections in ICU patients. This integrated multi-disciplinary approach will enhance the evidence for guideline development, thereby directly improving the health care response to the unprecedented threat of MDR-GNB at the local, regional and European level.

The results from R-GNOSIS will provide the following impacts:

- Improving management of Gram negative multi-drug resistant infections in the community as well as in health-care settings
- Improving our understanding of transmission and detection to decrease infection rates
- Improving management of Gram negative infections to retard the development of multi-drug resistance.

Public Health impact

Results of the R-GNOSIS project could be generalised and applied to settings and healthcare institutions in other European countries. Practical recommendations might be used by others, after taking into account differences in baseline resistance rates, patient case-mix and standards of care. Furthermore, this program should provide a sound basis for improved diagnostics, antibiotic prescribing practices and infection prevention strategies in the future and may generate a direct impact on clinical practice and appropriate administration of broad-spectrum antimicrobial agents, replying thus to the goals recently communicated in the EU Council conclusions (10 June 2008) on Antimicrobial Resistance, in which the EU “stresses the need of research in the area of AMR, e.g. to increase the understanding of the mechanisms and underlying risk factors that advance the development of AMR and to increase the knowledge of the effectiveness of current and future control measures.”

Finally, R-GNOSIS will also reply to a top priority action item of the WHO's global strategy for containment of AMR (<http://www.who.int/drugresistance>) by "improving the use of antimicrobial drugs", since the comprehensive knowledge provided by R-GNOSIS will be directly used to improve antibiotic usage policies. Thus, the results of this project will be of value to policy makers, by providing data useful for evaluation of different national antibiotic stewardship and infection prevention strategies. Assessment of the usefulness of the recommendations derived from the results of R-GNOSIS should also be of valuable help for those in charge of infection control, for hospital administrators, and for those managing budgets of large healthcare organisations. Ultimately, this work will contribute to the prevention of antibiotic-resistant infections and improve quality of patient care in Europe.

Providing an infra-structure for further studies

With 5 international clinical intervention studies, enrolling close to 60,000 patients, yielding >100,000 clinical specimens for microbiological screening and thousands of MDR-GNB isolates R-GNOSIS will provide a formidable infra-structure for further studies. R-GNOSIS is – intentionally - almost completely focussed on Enterobacteriaceae producing ESBLs and/or carbapenamases. Naturally, the resistance issue in Gram-negatives is larger and can be extended to other species, such as *Pseudomonas aeruginosa* and *Acinetobacter* species, and other resistance mechanisms such as ampC production, porin expression and efflux pumps. The studies performed, the samples obtained and isolates collected will enable indepth investigation of other species and other resistance mechanisms, and the partners involved will actively seek additional funding (and collaborations) to further explore this enormous wealth of samples and data that will emerge from R-GNOSIS.

1.4.2 Dissemination and exploitation of the project results

The dissemination and communication about the project and its findings to the relevant target audiences is crucial for the success of the project. An information dissemination plan was set up at the start of the project, which detailed the project dissemination and communication activities, their objectives, targets, tools and associated budget. The objectives of the R-GNOSIS dissemination and communication activities were to divulge knowledge generated by the project and to promote scientific and technical progress on the efficacy and effectiveness of cutting-edge interventions to reduce acquisition, carriage, infection and spread of Multi-Drug Resistant Gram-negative Bacteria (MDR-GNB) to the scientific community and the stakeholders in infectious diseases, public health, policymakers, drug manufacturers, and health care workers. It also aimed to inform the public about ongoing research activities and their possible impact on society.

Communications

Communication was fundamental to R-GNOSIS since awareness of the results from R-GNOSIS is one of our major concerns. To this end a number of communication and dissemination activities were carried out. For instance, a fully functional and user friendly website was developed and maintained throughout the duration of the project and will even be maintained beyond it (at least 2 years after the end of the project). This web site includes information about the vision, objectives and outcomes of R-GNOSIS.

Publications

A large number of publications were presented both at conferences and in journals based on the results of the project. The major papers reporting results from the project were and will be published in peer-reviewed, prestigious scientific journals. Most of the partners already have an established tradition for joint publications in high impact journals. Furthermore, the results were and will be presented at national and international conferences (ECCMID, ID-week, ICPIIC, etc.) depending on the stage of the project.

Exploitation of foreground

The results of the project will allow all participating institutions to pursue internationally competitive high-quality research and facilitate future collaborative projects.

The project results will significantly advance the general knowledge for controlling the spread of MDR-GNB. WP2 has provided strong evidence that using the Flexicult™ test for women with UTI in primary care does not contribute to improving the quality of antibiotic prescription. Unfortunately final conclusion cannot yet be drawn from the other 4 clinical studies. Yet, for each of these studies patient enrolment has been finalized (with targeted patients numbers reached) and all partners involved agreed to compete analyses after the funding period. This was preferred over a scenario of early termination of patient enrolment, as this would jeopardize the scientific validity of studies because of insufficient patient numbers for meaningful analyses. The final results of these studies are expected in the fourth quarter of 2017.

The final findings of the clinical studies will be accompanied by in-depth studies on the bacterial ecology and on modelling studies providing support for clinical decision making in infection prevention.

1.5 Project partners and contact & project website

1.5.1 R-GNOSIS partners

The R-GNOSIS research program involved patients and clinicians, microbiologists and epidemiologists, infectious disease specialists, caregivers in inpatient care and mathematical modellers. The R-GNOSIS consortium was made of 24 partners from 9 countries which include Belgium, Denmark, France, Germany, Israel, Netherlands, Spain, Switzerland and United Kingdom.

- UMC Utrecht, Utrecht, Netherlands – Marc J Bonten
- Université de Genève, Geneva, Switzerland – Stephan Harbarth
- University of Antwerp, Antwerp, Belgium – Herman Goossens/Surbhi Malhotra
- TASMC, Tel Aviv, Israel – Yehuda Carmeli
- Department of Health, Public Health England, London, United Kingdom – Julie Robotham
- Université Paris XII – Val de Marne, Paris, France – Christian Brun-Buisson
- Université de Fribourg, Fribourg, Switzerland – Patrice Nordmann/Laurent Poirel
- Cardiff University, Cardiff, United Kingdom – Chris Butler / Tim Walsh
- University of Southampton, Southampton, United Kingdom – Paul Little
- Spanish Society and Family Medicine, Barcelona, Spain – Carl Llor
- Charité - Universitaetsmedizin Berlin, Berlin, Germany – Petra Gastmeier / Sonja Hansen / Friederike Maechler
- Da Volterra, Paris, France – Florence Séjourné
- Servicio Madrileño de Salud, Madrid, Spain – Rafael Cantón Moreno / Patricia Ruiz
- Statens Serum Institut, Copenhagen, Denmark – Niels Frimodt-Møller
- University of Oxford, Oxford, United Kingdom – Ben Cooper / Esther van Kleef
- ARTTIC, Paris, France – Carlos Triay / Andrea Kuperberg
- Check-Points Health, Wageningen, the Netherlands – Pieter Vos
- GENEWAVE, Paris, France – Yann Marcy
- Assistance Publique Hôpitaux de Paris, Paris, France – Antoine Andremont / Bruno Fantin
- HUMFRYX, Barcelona, Spain – Josep M Cots

1.5.2 Contact details

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1.5.3 R-GNOSIS project public website

A public website on the R-GNOSIS project is available at: <http://r-gnosis.eu/>

The website contains an overview of R-GNOSIS, more detailed information on its objectives, innovative aspects and work structure, as well as the research domain and main applications, the groups involved and their contributions, downloadable publications and links to other related information sources on the web and a contact email. The publications and outputs pages are regularly updated, as well as the Events & Meetings page for the internal and external communication of the project as widely as possible.