agence d'évaluation de la recherche
et de l'enseignement supérieur

Section des Unités de recherche

AERES report on the research unit

Laboratoire d'Ingéniérie des Systèmes Macromoléculaires

From the

Université de la Méditerranée

CNRS

January 2011

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Université de la Méditerranée

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Le Président de l'AERES

Didier Houssin

Section des unités de recherche

Le Directeur

Pierre Glorieux

January 2011

Research Unit

Name of the research unit: Laboratoire d'Ingéniérie des Systèmes Macromoléculaires

Requested label: UPR

N° in the case of renewal: 9027

Name of the director: M. James STURGIS

Members of the review committee

Committee chairman:

Ms. Françoise JACOB-DUBUISSON, CNRS, CIIL, France

Other committee members:

M. Jean-Marie RUYSSCHAERT, Université libre de Bruxelles, Belgique

M. Arnold DRIESSEN, Groningen University, The Netherland

M. Ian HENDERSON, Birmingham University, United Kingdom

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M. Jean-Claude PORTAIS, INSA Toulouse, France

Ms. Pascale ROMBY, (CoNRS), Strasbourg, France

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M. Yves GAUDIN

University, School and Research Organization representatives:

M. Pierre CHIAPPETTA (University of Aix-Marseille 2)

M. Gilbert DELEAGE (CNRS)

Report

1 • Introduction

- **Date and execution of the visit :**

The visit took place on January 31st, 2011 and was carried out by an international team of 7 qualified scientists with complementary expertise in the research areas of the 6 teams evaluated. The visit started with a general presentation of the laboratory by its director, its history and achievements and the past and future organizations. The team leaders presented their results and projects and answered to questions of the committee. Committee members met with the CNRS and University Aix-Marseille 2 representatives, the PhDs and postdocs, the technical staff and the scientists. After a final meeting with the director of the laboratory, the committee gathered on February 1st, 2011 to establish the present report.

- **History and geographical localization of the research unit, and brief presentation of its field and scientific activities :**

This laboratory is located on the campus of the Institut de Microbiologie de la Mediterrannée in Marseille. The LISM was initially founded in 1992. Since its inception the Unit has worked on the structure and dynamics of protein complexes in the bacterial envelope and on their engineering for biotechnological applications. Upon the departure of the group leader 3 years a new leader took the job. The large Pseudomonas group previously led by the unit head was split into two smaller groups in 2009 under the impulse of the new director. The laboratory has undergone additional changes recently. In particular one research group has left for another campus, and new personal has been recruited. The unit has been organized into 5 research groups for the last 2 years. It is proposed to split one of the groups to establish a new group in the next 4 years.

During the years the unit has kept the same focus on biological membranes and in particular on multiprotein complexes found in the envelope of Gram-negative bacteria. Their emphasis is on fundamental reasearch. Subjects of interest are protein secretion, membrane protein assembly and dynamics, sensing and interaction with the environment, metabolism, macromolecular import and community lifestyles. Some of their model organisms are pathogens, but the emphasis is not placed strongly on pathogenesis. To address their scientific questions the unit has developed a combination of approaches including microbiological, biochemical and computational techniques, spectroscopy, microscopy, interactome and transcriptome analyses.

- **Management team :**

The Unit is directed by a senior member who also leads one of the 5 groups that currently compose the Unit. The groups leaders hold regular meetings together to discuss organizational questions within the unit.

- **Staff members (on the basis of the application file submitted to the AERES) :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)	4	5
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)	9	10
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)	5	
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)	10	10
N5: Number engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)	2	
N6: Number of Ph.D. students (Form 2.7 of the application file)	12	
N7: Number of staff members with a HDR or a similar grade	8	8

2 • Overall appreciation on the research unit

- **Summary :**

The LISM is a small-size research unit focused on fundamental questions related to the bacterial envelope, and in particular on large protein assemblies and machineries in the Gram-negative envelope. It contains some solid groups that perform good quality research. The researchers of the unit have a strong and recognized expertise on the Gram-negative envelope. The unit as a whole shares a number of tools and ideas, but the committee has noted some lack of a long-term strategy to optimize the full potential of the unit and its member groups.

- **Strengths and opportunities :**

The LISM operates in a good environment, given the large community of microbiologists on campus. The expertise of the unit members on the Gram-negative envelope is internationally recognized. The topics and objectives of the various groups are rather homogeneous. They have developed and share a set of useful tools, including a pathogen-host interaction platform based on the C. elegans model which is used by several groups of the unit.

The age profile of the unit, which is mostly composed of young researchers is clearly an asset. The member groups have had a good success at fund raising, essentially from national sources. The unit has also been able to recruit talented young people during the last years. The strong links of the unit with the University have also enabled it to attract several good PhD students.

- **Weaknesses and threats :**

The unit appears to need a stronger global sense of direction. A well-defined long-term strategy will be necessary to strengthen its visibility durably. The team of the director works on issues that are somehow peripheral relative to the preoccupations of the rest of the unit and is focused on different models. This may contribute to the lack of a global strategy. There are clear opportunities to foster stronger and new interactions between the teams. They would be instrumental to enhance the unit coherence and scientific leadership.

The unit as a whole but in particular the smaller of its groups might risk to go under the critical size to maintain competitiveness, especially given the dispersion of some groups on a multitude of projects. This is also the result of a

significant lack of space, which threatens the development of the unit such as the recruitment of new groups or the expansion of the smaller ones.

Very few contacts exist between the unit and industry, and the unit does not attempt to develop applications.

- **Recommendations :**

The committee recommends that a global and more integrated strategy be defined for the evolution of the unit in the coming years. This is a crucial aspect so that the unit could benefit from the planned scientific reorganization of the CNRS campus in the near future ; it is certainly an opportunity to claim more space. The younger group leaders should be helped to refocus their priorities in order to improve their visibility and sustainable productivity. The management team should set productivity targets for the unit, the group leaders and the PhD students.

The unit should integrate new technical developments in their projects. This implies defining their needs and engaging in discussions at the level of the institute to set up strategies for exploiting the existing facilities in an optimal way and for developing new ones.

The international visibility of the unit and its member groups has to be enhanced, for instance by applying for European grants.

A redistribution of the technical staff –particularly in the view of the creation of a new group- should be envisaged as the current distribution is uneven.

- **Production results :**

(cf.http://www.aeres-evaluation.fr/IMG/pdf/Criteres_Identification_Ensgts-Chercheurs.pdf)

A1: Number of permanent researchers with teaching duties (recorded in N1) who are active in research	4
A2: Number of permanent researchers without teaching duties (recorded in N2) who are active in research	9
A3: Ratio of members who are active in research among staff members [(A1 + A2)/(N1 + N2)]	1
A4: Number of HDR granted during the past 4 years	3
A5: Number of PhD granted during the past 4 years	7

3 • Specific comments

- **Appreciation on the results :**

The research performed in the various groups is at a good level, although no major breakthroughs have been achieved in the recent years. Their scientific productivity is also rather good, with an average of 1-2 papers per year and per person, mostly in leading international journals in their field. A fair proportion of these publications involve more than one group. Several of the articles in the higher-ranking journals are the result of collaborations, in particular with the previous unit leader. The average number of citations after two years of publication is 13.5. Since 2006, the unit has produced 1 patent, a few book chapters and 93 papers. Of these papers, in 54 cases, members of the unit are positioned as first and/or senior authors.

The unit has published 14 articles in J. Bacteriol (IF=3.94), 6 in the Journal of Biological Chemistry (IF=5.32), 5 in Molecular Microbiology (IF=5.36), 4 in Biophysical Journal (IF=4.39), 4 in the Journal of Molecular Biology (IF=3.87), 4 in Biochemistry (IF=3.22), 3 in PNAS (IF=9.43), 3 in Environmental Microbiology (IF=4.9), 3 in Proteomics (IF=4.42), 3 in Infection and Immunity (IF=4.2), 3 in the Journal of Structural Biology (IF=3.67), 2 in Photosynthesis Research (IF=2.3), 2 in Applied Environmental Microbiology (IF=3.68), 1 in Microbiology and Molecular Biology Reviews (IF=12.58), 1 in EMBO Journal (IF=8.99), 1 in EMBO reports (IF=6.9), 1 in PLoS Pathogen (IF=8.97).

A number of 8 PhD theses and 3 HDR theses have been defended during the reporting period of 4 years.

- **Appreciation on the impact, the attractiveness of the research unit and of the quality of its links with international, national and local partners :**

As a whole, the number of invitations to high-level international meetings is not remarkably high, although some individuals have a better visibility (see group by group analysis below). Altogether, there have been 11 invitations to prestigious meetings or conferences (Gordon, CSH, EMBO, ASM, FEMS, ESF-EMBO, IUMS). This moderate record could be explained by the young age of most group leaders.

The attractiveness of the unit is good. Thus, 2 young researchers who originate from other labs in France have recently joined the unit. In addition, the unit has recruited one researcher and has obtained one Chaire d'Excellence University-CNRS. All the new recruits are French.

The member groups have been quite successful in raising national financial support. Several young leaders have obtained an ANR Jeune chercheur grant, and the others have obtained or participate in ANR grants as well (12 ANR grants in total for the reporting period). Additional sources of funding are the FRM and VLM foundations. Some of the groups participate to ERA-NET programs, in one case as the coordinator. In contrast, they are not involved in major EU programs nor have they received significant industrial funding.

All the groups participate in several national and international networks and collaborate with partners at the global scale.

- **Appreciation on the management and life of the research unit :**

The unit is composed of 5 groups (6 in the near future), and a stated goal is to keep them all at similar sizes in order to avoid marked imbalances between them. While each individual group needs to be large enough to be competitive, it is unsure that it will be tenable to limit the development of some of the groups in the longer term. At this moment expansion is not possible because of the lack of space. This is the major complaint in the unit. A good atmosphere appears to exist, and the day-to-day organization seems to be satisfying to all.

The management team is composed of the group leaders, who meet 4-5 times a year to stimulate internal collaborations and facilitate synergy between groups. It appears that communication should be improved between groups. Students in particular are not necessarily aware of the research done in the other groups. At this moment, only 4-5 unit research meetings are organized per year; this number should be raised.

Similarly, setting up a Journal club in the unit would give students the opportunity to present and discuss published work and broaden their scientific culture.

The involvement of the unit in the animation of research at the local level is significant. A member of the unit organizes students' seminars for the entire institute on a weekly basis.

The unit has strong connections with the university. Thus, 5 members of the unit assume teaching duties as professor (1) or associate professors (4), and a few other researchers of the unit are engaged in teaching too.

- **Appreciation on the scientific strategy and the project :**

The committee's analysis is that the unit should establish a more ambitious, global strategy exploiting better the technical and scientific relatedness between the projects of the various groups. This sharing of tools, ideas and of some facilities appears to be the main added value of the unit as a whole.

A stronger collective leadership might prove essential to define the strategic directions of the unit and to enhance its visibility. This will be particularly important to increase the size of the unit and to plan for new, centralized facilities when the reorganization of the campus is discussed in the near future.

The committee has also noted the need for new technical developments to progress in the systems under study. Cryo-electron tomography and state-of-the-art fluorescence imaging techniques would be an asset. It would be necessary to discuss which additions are needed to reinforce the unit. Some of them in particular may demand the recruitment of new groups of specialists.

Regarding the allocation of human resources, the uneven distribution of the technical personal between groups needs to be addressed.

4 • Appreciation team by team

- **Title of the team and team leader :**
- **Group E1: Secretion systems and pathogenicity in Pseudomonas aeruginosa**
- **Leader: Romé VOULHOUX**
- **Staff members :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)	1	1
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)	2	2
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)	0	
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)	2	2
N5: Number of engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)	0	
N6: Number of Ph.D. students (Form 2.7 of the application file)	4	
N7: Number of staff members with a HDR or a similar grade	2	2

- **Appreciation on the results :**

The research in this group aims at the discovery and analysis of various secretion systems and secreted effectors in Pseudomonas aeruginosa with a link to pathogenesis. Research is subdivided in five main topics, i.e. T2SS, T3SS, T5SS, T6SS and the relation between TAT and virulence. The major results of the work during the reporting period have been the elucidation of the NMR structure of the soluble domain of the major pseudopilin of the T2SS and a model for the pseudopilus assembly. The TAT secretome has also been explored. A hybrid T5S system has been identified. In collaboration with the former head of the unit multiple T6SS have been identified. The focus in T6SS is on regulation and on the identification and function of effectors.

Each of these topics strongly depends on external collaborations. There are also interactions within the unit, and continued interactions with the former group leader.

The group seems to be in a transitional phase due to the departure of the former group leader. Therefore, the exact output cannot be fully assessed. In recent years, the new group leader has started to publish and to obtain grants independently.

The work is of good quality and has been published in general biochemistry and microbiology journals. However, most of the manuscripts are the result of collaborative efforts. Thus, out of a total of 23 papers, group members have signed in first or last position in only 8 of these : 2 J Bacteriol, 2 JBC, 1 Microbiology, 1 Appl. Env. Microbiol, 1 Env. Microbiol., 1 Int. J. Med. Microbiol.

The impact of the group is not as high as compared to peers working on similar topics, likely because of the large number of topics for a relatively small team. Therefore, the committee noted that it might be difficult to be internationally competitive at the highest level. To maximize output, the group is involved in a wide network of collaborations in particular on structural aspects of the work. This is done very efficiently, but there is a concern that credit for this work eventually will go to the collaborating party and not the research team.

Two PhD theses were defended in this group during the reporting period.

- **Appreciation on the impact, the attractiveness of the team and of the quality of its links with international, national and local partners**

The group is headed by a young and enthusiastic researcher and is supported by a senior assistant Professor. The team is clearly capable of recruiting good scientists (a CNRS CR1 researcher position was recently attributed), and this is to the credit of the enthusiasm and interest of the leaders. The attractiveness of the team is further reflected by the fact that they have recently recruited 2 PhD students, and that one 2nd year and one 3rd year students complete the team.

The team leader has been invited for one talk and has helped to organize one international conference. This is rather low for a team leader with a group of this size.

The group has been quite successful in recent years in obtaining scientific grants from various national granting agencies, and it participates to ERA-Net and Marie Curie international scientific networks. It has also attracted post-docs, some of them foreigners. This is very good for a young team and reflects the level of their scientific ambitions.

The team is involved in a wide range of international collaborations spanning three continents. Whether these are stable collaborations is difficult to judge but it is hard to envisage that the team will be able to maintain 8 high-level collaborations in the diverse research areas. One industrial contract has been obtained (technology transfer contract).

- **Appreciation on the scientific strategy and the project :**

The team proposes an ambitious plan that concerns a continuation of current research, with a strong emphasis on structural analysis and the identification/elucidation of virulence factors. Because many objectives are being pursued, the program relies on a large number of international collaborations with top scientists in structural biology and pathogenesis. The work can be of high impact when successful and it will concern cutting-edge work, but this will be at the expense of the visibility of the group.

The group is equipped with a wide range of expertise that can support these projects, which is a strength. However, this diversity of projects is unsustainable, and the team needs to focus much more. They must identify a select number of topics and focus upon these topics to ensure they gain and maintain a niche and an international identity in the field. Importantly, they must limit their activities to ensure they are not outcompeted by other groups in the world.

- **Conclusion :**

— Summary :

This group works on protein secretion in Pseudomonas aeruginosa and the link between secretion and pathogenesis. A number of subjects are being addressed in addition to the initial core focus of the group on the

T2 secretion machinery, including the identification and function of secreted effectors, the characterization of a new subtype of T5S, and the TAT system. This large dispersion has led the group to set up international collaborations with specialists in structure and eukaryotic cell biology.

— Strengths and opportunities :

The group is quite young and highly motivated. They have just recruited a new permanent researcher who has a good expertise in protein secretion. Their financial support is good. The organism under study is an excellent model to address question in secretion and pathogenesis. In addition, its importance as a pathogen should provide opportunities to team up with medical researchers and eventually contribute to drug development strategies. The group has excellent international collaborations in particular with structural biologists that likely will increase the impact of the work.

— Weaknesses and threats :

The lack of focus is a major concern; too many systems are tackled simultaneously with little critical mass on each topic, while a strong international competition exist on most of the topics developed by the group. The productivity of the team has been rather low, with no high impact papers. Most topics strongly depend on external collaborations, in which the group will not have the lead.

— Recommendations :

The group should focus essentially on the mechanistic aspects of T2S. There might be a good opportunity given that the field appears to be less crowded than it has been in the past. In addition, if they want to really make breakthroughs in the function of secreted protein, they should team up with medical researchers to study the pathogenesis related processes.

- **Title of the team and team leader :**
- **Group E2: Dynamics and assembly of membrane proteins**
- **Leader: James STURGIS**
- **Staff members :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)	2	2
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)	2	2
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)	2	
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)	1	1
N5: Number of engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)	0	
N6: Number of Ph.D. students (Form 2.7 of the application file)	2	
N7: Number of staff members with a HDR or a similar grade	2	2

- **Appreciation on the results :**

The research of this group aims at a molecular understanding of the interaction between peptides and proteins within biological membranes. Special attention is given to a molecular description of the interaction between single transmembrane domains by coupling experimental and theoretical tools along the lines of the pioneering work of Engelman. Convincing effort is made to decipher the code that links sequence to structure in membrane proteins. There is a real willingness to test the validity of the biophysical data in terms of biological activity. AFM data obtained in collaboration with the Institute Curie provide important information about the organization of proteins within the photosynthetic membrane system.

The work is of good quality and internationally recognized. It is performed in collaboration with national and international partners yielding a good input with publications in good-impact journals. 36 publications have been signed by group members in the reporting period. In these they are first or last author(s) in 12 of them including 1 PNAS, 1 J Struct Biol, 1 Biophysical J., 1 J Mol Biol, 1 Biochemistry, 2 J Phys Chem, etc. Some of their other publications in high impact journals result from collaborative work (Oncogene, Mol Biol Cell etc.).

- **Appreciation on the impact, the attractiveness of the team and of the quality of its links with international, national and local partners**

The group has strong roots in experimental and theoretical analysis of membrane protein folding and complex formation despite its relatively small size. It is well organized and has successful international collaborations. It has succeeded in recruiting a young permanent researcher recently.

The group leader is known as a very good spectroscopist, and the expertise of the group on photosynthesis apparatus is internationally recognized. However, the group leader is rarely invited for international meetings, but he has been

successful in obtaining national grants (e.g. 2 from ANR). There is also a participation in a EU-EST program. There are no clear industrial links or links of medical importance.

The basic research performed by this group does not interconnect well with the other groups of the unit trying to understand membrane protein assemblies in the Gram-negative envelope. Therefore, there are few internal collaborations although the other groups might benefit more from the knowledge of this group on polytopic membrane proteins.

- **Appreciation on the scientific strategy and the project :**

The project for the next 5-years is a continuation of the work carried out during the previous period and will address the main questions raised previously regarding the helix-helix interactions, lipid-helix interactions, membrane protein folding and membrane organization that have been classically investigated in the group. There will be an intensification of the molecular dynamics work to study membrane protein structure, although it would be advised to include experimental work to validate the predictions.

Fluorescence techniques are used successfully to characterize at a molecular level the homo-and hetero interactions of synthetic peptides mimicking the transmembrane domains of receptors. X-ray crystallography would provide additional information about the structure, the distance and the organisation of the TM helices.

Altogether they have a solid project that matches well their expertise.

- **Conclusion :**

— Summary :

This group performs solid scientific work in the fields of membrane protein folding and transmembrane helix packing using a combination of theoretical and experimental approaches. They have a highly integrated work program and they are recognized in their domain. The committee has raised the issue of their connections with the rest of the unit.

— Strengths and opportunities :

The group has a well-structured project, with a good link between experiment and theory. It has a significant experience with diverse biophysical techniques. It is visible nationally and internationally from the work on the organisation of the photosynthetic apparatus in photosynthetic bacteria. They also have excellent national and international collaborations.

— Weaknesses and threats :

The work strongly depends on interactions with others, in particular on a very productive collaboration with AFM specialists. The group is rather small and has produced few theses. It lacks links with the pharmaceutical industry. The leader is not involved in editorial work and has limited EU visibility. The group collaborates little with the rest of the unit because of its own focus.

— Recommendations :

The group should seek to establish internal collaborations for instance by the development of techniques that could benefit other teams in the unit and by addressing the biological systems under study in the other groups. In order to progress in their studies they should consider including single-molecule fluorescence spectroscopy methods to analyze membrane protein conformation and dynamics. Also X-ray crystallography studies carried out in collaboration with other groups in Marseille would not only contribute to a deeper molecular understanding of the interactions between transmembrane domains but would also benefit the other teams involved in the purification and expression of bacterial membrane proteins.

- **Title of the team and team leader :**
- **Group E3: Macromolecular transport through the bacterial cell envelope**
- **Leader: Roland LLOUBES**
- **Staff members :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)	0	0
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)	3	2
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)	1	
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)	2	1
N5: Number of engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)	0	
N6: Number of Ph.D. students (Form 2.7 of the application file)	3	
N7: Number of staff members with a HDR or a similar grade	2	1

- **Appreciation on the results :**

The team has pursued their internationally recognized research on the Tol system and on the mechanism of action of colicins. Recently, work was also developed on the type VI secretion system (T6SS) by a researcher who will establish a new team on this topic at the LISM.

During the reporting period, a major effort was dedicated to establish the interactions, assembly and motions of transmembrane segments of TolR and TolQ. The work provides a number of contributions towards understanding assembly of the TolA-Q-R complex. Work on colicins has focused on the mechanism of action of endonucleasic colicins and on the interaction between colicins and their immunity proteins. Novel findings were obtained for both processes. Studies have been carried out on the T6SS using the genetically tractable enteroaggregative Escherichia coli (EAEC) as a model system.

The team showed a steady production of publications on its research themes that came out in leading journals of the field. In most of their work published during the reporting period they signed as leading authors : 5 J

Bact, 3 JBC, 1 Mol Microbiol, 1 J Mol Biol, 1 Biochemistry. The international recognition of the team in its fields of research is attested by reviews on colicin biology and on T6SS published in high-impact journals (EMBO Rep, MMBR).

3 PhD theses were defended in 2010.

- **Appreciation on the impact, the attractiveness of the team and of the quality of its links with international, national and local partners**

The permanent researchers of the team were invited to give conferences abroad and to participate in national and international committees. A young member of the team has been particularly active, being namely member of the editorial boards of two leading microbiology journals.

The group has been very successful at raising fund form national sources (2 ANR, CNRS-PEPS, VLM and FRM). A productive engineer who worked previously on TonB joined the team recently.

The new T6SS subject has attracted several students, one post-doc and a permanent researcher, who will all be instrumental to develop this theme competitively.

The Ton/Tol project is being pursued in good strategic collaborations with several highly recognized structural biology groups and with an expert team on cell wall analysis to characterize the enzymology of colicin ColM. The stuctural work will likely prove essential for detailed molecular insight on the mechanisms studied by the team. The research activity has the potential to translate into applied tools to control bacterial growth and pathogenesis.

- **Appreciation on the scientific strategy and the project :**

The Tol/Ton project will be the continuation of previous work, with a marked diversification of the approaches and the development of pertinent collaborations. In particular, the structural studies look promising (diffracting crystals have been obtained) and could provide them with important information for further mechanistic studies. It appears that the group has taken the lead in this collaboration. The newly recruited engineer has the perfect expertise to pursue this work, as attested by his own publications on TonB. Some projects like the crystallization of the whole TonB complex are risky but deserve to be done.

Another collaboration has been established to reconstitute and study the organization of the inner membrane Tol and Exb complexes. A new Vibrio phage model will be set up. The colicin projects will be developed with a focus on dimeric organization of immunity proteins and on ColM.

Research on the T6SS will be continued in an independent research group (E6).

- **Conclusion :**

— Summary :

This is a solid group that is doing excellent work. The project will be pursued by diversifying approaches, in particular with a strong collaboration with a top crystallography group in the US. The work performed on colicins is also well recognized will be pursued using several model systems, with a strong emphasis on structure-function relationship.

— Strengths and opportunities :

The group has an extensive expertise on the Tol and colicin systems. It has been reinforced recently by the recruitment of an engineer who is a specialist on this topic. Collaborations with excellent teams working on the structural biology of bacterial envelope proteins and with a highly recognized specialist on cell wall biogenesis and structure are significant assets for the projects. With 3 permanent researchers, the group has a good potential for training people.

— Weaknesses and threats :

The group is facing a strong decrease in size and will need to recruit new students. They should evaluate the added value of the CTX phage project, with respect to the distribution of human resources.

— Recommendations :

The group should exploit the generated structural biology information to design investigation on structure-function correlation. They are also opportunities to diversify their experimental approaches (dynamics using fluorescence techniques, cryo-EM analyses to study colicin entry into cells). Links should be kept with the new group.

- **Title of the team and team leader :**
- **Group E4: Protein complexes and bacterial metabolism**
- **Leader: Emmanuelle BOUVERET**
- **Staff members :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)	0	1
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)	1	1
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)	2	
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)	0	0
N5: Number of engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)	1	
N6: Number of Ph.D. students (Form 2.7 of the application file)	2	
N7: Number of staff members with a HDR or a similar grade	1	1

- **Appreciation on the results :**

The research developed by this group aims at getting a better understanding of the mechanisms of stringent response in Escherichia coli. More particularly, the group focuses on the relationships between stringent response and lipid metabolism. This positioning is original and relevant since most of the current knowledge on stringent response is related to amino acid starvation. During the reporting period, the group has investigated the interactions between the enzymes responsible for (p)ppGpp synthesis and degradation and components of lipid metabolism. In particular, the team has shown that SpoT interacts with the acyl carrier protein ACP, a key component of lipid biosynthesis, making a clear link between stringent response and alterations of lipid metabolism. Conversely, the group has also investigated the control of lipid metabolism in response to stress using genetic approaches, and they have identified critical control points. The second project deals with the protein-protein interaction network involved in lipid metabolism. They have improved and developed new tools -new 2-hybrid vectors and protein tagging techniques for tandem-affinity purification (TAP)- to study protein/protein interaction networks.

The group has an impact in the fields of microbial physiology and proteomics. One of their papers on the TAP approach in particular is highly cited. The group has published 6 articles between 2006 and 2010 in good microbiology and biochemistry journals : 2 J. Bacteriol., 1 Mol. Microbiol. and 3 Proteomics. During the same period, the group has also contributed to 2 book chapters. Two PhD theses were completed during the reporting period.

They collaborate with other teams of the LISM, in particular within an ANR program led by the director of the unit, which resulted in a common publication.

- **Appreciation on the impact, the attractiveness of the team and of the quality of its links with international, national and local partners**

The financial support of the team is good and the international recognition of the leader, although rather recent, is attested by her invitation to 3 international meetings (2 Gordon conferences) and the invitation to writing a review for the well-known book "The stringent response" edited by G. Storz. Although the group has a limited size, the group leader got 2 ANR (1 ANR to support young researcher's project) from which one post-doc and one engineer have been recruited. The team also received specific support from the CNRS through a PEPS grant. The team is well integrated

into European consortium since one of the PhD students was recruited in the frame of an Initial training of researchers. A "Chaire d'excellence" CNRS-University was recently attributed to a new recruit of the group.

- **Appreciation on the scientific strategy and the project :**

The project for the next 5-years period is a continuation of the work carried out during the reporting period, and represents further investigation of the stringent response in E. coli and its relationships with lipid metabolism. It includes two main aspects: firstly, understanding of how lipid stress induces the stringent response will be studied using in vitro and invivo approaches; secondly, the team will investigate the protein-protein interaction network that ties together lipid metabolism and ribosome biogenesis and the genetic regulation of lipid biosynthesis in response to stress. The role of small non coding RNAs in this process will also be addressed in collaboration with a researcher recognized in the field.

The observations made with E. coli will be extended to Salmonella typhimurium for generalization purpose but also to investigate the role of these mechanisms in pathogenicity. The expertise of the newly recruited researcher in the group fits with the new model.

This is a well-constructed project, which has received good financial support. However, it is quite significant compared to the size of the group and it will need the application of technologies that are novel to the group. This will be essential in order to keep their edge in this competitive field.

- **Conclusion :**

— Summary :

The team has now acquired an international visibility on the bacterial stringent stress and growth control. The projects are suitable to achieve long-term goals toward the determination of the links between lipid biosynthesis and ribosome biogenesis in response to the stringent control. The group has a good niche but its small size may be limiting for the development of their project.

— Strengths and opportunities :

This is a young group that is taking off. The originality of their project will lead to a good international visibility. They have set up an original method to analyze protein networks at the membrane and involved in lipid biogenesis. The collaboration with a new team working on ncRNA regulation will be an added value to the proposed project and will certainly enhance their competitiveness. Financial support is good.

— Weaknesses and threats :

The small size of this team and the absence of clear plans for further expansion represent potential risks of being overtaken by international competition. It seems that the extension to the Salmonella model is the result of a scientific strategy and not of a project designed for the new researcher, but the team has to be aware to avoid dispersion.

— Recommendations :

The team has a small size but collaborations with external teams or from the unit would certainly benefit the project and help the team to increase the list and the quality of their publications. This aspect has already been taken into account since the team has started an external collaboration to work on RNA regulation. More technical support would also be welcome.

- **Title of the team and team leader :**
- **Group E5: Sensing the environment and community lifestyle in Pseudomonas aeruginosa**
- **Leader: Sophie DE BENTZMANN**
- **Staff members :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)	1	1
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)	1	1
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)	2	
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)	0	0
N5: Number of engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)	1	
N6: Number of Ph.D. students (Form 2.7 of the application file)	3	
N7: Number of staff members with a HDR or a similar grade	1	1

- **Appreciation on the results :**

This team results from the split of the large Pseudomonas group led by the former unit head. It develops two topical and interconnected projects dealing with molecular machines located at the cell surface of Pseudomonas aeruginosa and the identification of regulatory networks that control the transition between planktonic and sedentary (biofilm formation) lifestyles. The first project has led to the characterization of genetic loci encoding chaperone and usher proteins involved in the assembly of fimbriae, and proteins involved in the assembly of adhesive type IVb pili. In the frame of a EURANET-ANR, the team has developed an "adhesive" chip with the aim to understand biofilm-associated antibiotic resistance mechanisms. For the second project, using a combination of approaches (genetic, transcriptomic, two-hybrid system), three new signaling pathways have been identified to control the transition to biofilm formation. These signaling pathways converge to the synthesis of two non-coding RNAs (ncRNA) that regulate gene synthesis at the post-transcriptional level. Finally, Caenorhabditis elegans has been used to identify virulence factors of P. aeruginosa isolated from cystic fibrosis patients.

The team has a good publication record (20 publications) including one in EMBO J as leading author and several articles in the best journals of microbiology, including 4 J. Bact, 1 Mol Microbiol, 2 Env. Microbiol. The other publications of the group result from collaborations, notably with the former group leader but also with other partners. One patent has been obtained for the microarray. The team actually hosts three PhD students and 2 post-docs.

- **Appreciation on the impact, the attractiveness of the team and of the quality of its links with international, national and local partners**

The group is in a transitional period following the restructuration of the larger Pseudomonas team. They are becoming internationally visible as attested by the invitations of the permanent researchers to a few international meetings.

The external funding of the team is excellent. They coordinate an ERA-net program and have obtained several ANR grants and grants from foundations. For valorization, a patent has been obtained on the microarray design for analyzing regulatory networks involved in biofilm formation.

The group has attracted students and post docs. The group leader is dynamic and very active nationally in evaluation committees and coordinates a GDR on Pseudomonas. They are also involved in various national collaborations thanks to their expertise on Pseudomonas and the tools they have developed.

- **Appreciation on the scientific strategy and the project :**

The projects of the team will be pursued in the light of the data obtained during the previous period and will include collaborations with laboratories with complementary expertise. In the first topic, the team will characterize more precisely the structure-function of several molecular machines (Type IV pili, CU family fimbriae and lectins) involved in the development and structure of biofilms. Through collaborations with crystallographers and chemists they will try to determine the structure of various adhesins and to identify the sugar moieties to which they bind. They will also develop screening for anti-adherence molecules in collaboration with a local platform, and transcriptomic microarrays to decipher gene expression from P. aeruginosa infected animal models or from clinical human samples.

In the second topic, the team will focus on the mode of action of the ncRNAs, the identification of the signals triggering the identified two-component systems and the missing partners involved in the signaling pathways and regulation. It should be noted that this last project is developed in collaboration with the former leader. The visibility of the group in this project thus remains to be established independently. In contrast, the group has established an independent and leading position on the adhesin project.

- **Conclusion :**

— Summary :

The team is working in the fields of regulation of biofilm formation by Pseudomonas and the characterization of the adhesins and surface machineries involved. They have a good international visibility. The projects are suitable to achieve long-term goals toward the understanding of the regulatory networks involved in the transition from the plantonic to the community lifestyles of the human pathogen P. aeruginosa.

— Strengths and opportunities :

Pseudomonas is an excellent model system to study the regulation and the molecular determinants involved in biofilm formation. The group has developed good tools and their collaborations with external teams having complementary expertise will certainly benefit the project. Their financial support is good. The group leader is very dynamic.

— Weaknesses and threats :

The dispersion between so many projects creates a risk for international competitiveness. The independent positioning of the group relative to the lab of the former leader remains to be asserted. At present their strategic choice of topics remains unclear. The group has no permanent technical help.

— Recommendations :

The group should refocus their effort on projects that will enable it to gain recognition independently. There is a need to recruit in the nearest future a permanent full-time researcher and/or full-time permanent technical assistance.

- **Title of the team and team leader :**
- **Group E6: Assembly of trans-envelope complexes**
- **Leader: Eric CASCALES**
- **Staff members :**

	Past	Future
N1: Number of researchers with teaching duties (Form 2.1 of the application file)		2
N2: Number of full time researchers from research organizations (Form 2.3 of the application file)		2
N3: Number of other researchers including postdoctoral fellows (Form 2.2 and 2.4 of the application file)		
N4: Number of engineers, technicians and administrative staff with a tenured position (Form 2.5 of the application file)		0
N5: Number of engineers, technicians and administrative staff without a tenured position (Form 2.6 of the application file)		
N6: Number of Ph.D. students (Form 2.7 of the application file)		
N7: Number of staff members with a HDR or a similar grade		1

- **Appreciation on the results :**

The project by this team seeks to utilize the enteroaggregative E. coli (EAEC) T6SS to define the molecular assembly of this secretion pathway. T6S was discovered rather recently, and the molecular mechanisms of this complex machinery remain to be deciphered.

The team has chosen to work on a genetically tractable system whose results can be widely applied to understanding T6SS. In addition, their model is an important pathogen since EAEC are a major cause of pediatric diarrhea leading to significant mortality and morbidity in the developing world and the largest cause of community acquired sporadic diarrhea. Despite the identification and study of EAEC for the last 25 years the mechanisms of pathogenicity remains enigmatic.

The group is a new team whose leader was previously member of team E3 where it established his own theme of research. Important tools have already been setup, and the team has already begun to publish on this new topic.

- **Appreciation on the impact, the attractiveness of the team and of the quality of its links with international, national and local partners**

The team leader has an excellent output. He has a consistent track record of publication for the last 10 years with a cumulative 29 published articles having an average citation of 44 citations per article and a cumulative 1,275 citations giving an H-index of 17. This is very good for a young researcher starting his group. He has recently given 7 departmental seminars and 3 invited talks at international conferences indicating he has a high international profile. His recently published articles include reviews (see above), and research papers in leading journals of microbiology and biochemistry: J Biol Chem and Mol Microbiol. The other scientist within the team also has an excellent output in the area of macromolecular secretion structures.

The team leader initiated important collaborations with structural biologists to determine the structure of the T6SS components and with experts in regulation to determine how T6SS expression is controlled. Several of these collaborations are local and are expected to be stable and strong.

- **Appreciation on the scientific strategy and the project :**

The project aims to define the assembly and architecture of the recently identified T6SS using EAEC as a model system. Studies are proposed on the machinery organization at the bacterial envelope and on its extracellular portion, which resembles long phage tails. Research on regulation of the T6SS gene clusters expression is also pursued.

The research theme fits very well in the technical expertise and the field of research of the LISM. The new group leader has extensive expertise on Gram-negative envelope and secretion to carry out the research proposed. His previous scientific achievements and the funding rose for this project provide a good basis to carry it out. This field is highly competitive. Innovative research will require very good strategic choices on the questions to target and strengthening of the team. The collaboration with structural biology groups having expertise on large protein assemblies' production and characterization is a good choice to support competitiveness.

- **Conclusion :**

— Summary :

This is an excellent project by a team of young researchers with high international profiles. The model system is relevant, tractable and an important pathogen. Although the T6S field is competitive, it appears that most groups are investigating the function of T6S systems in various bacteria. The focus of the group on assembly and mechanisms of the machinery thus represents a good niche for now.

— Strengths and opportunities :

A timely project on a high-impact topic in the field of bacterial secretion and pathogenesis. The group leader has expertise and raised consequent funds to carry out the research proposed. The project has already attracted another CNRS researcher to join the team and several students and post docs. The project is supported by a good level of funding and well-chosen collaborations.

— Weaknesses and threats :

Research on T6SS is highly competitive and progresses very fast in different model systems. A threat is that bigger groups might move into the field.

— Recommendations :

The team should refrain from developing multiple unfocused projects that distract it from the central T6SS studies. It will need a very effective strategy to tackle original questions. It should also increase the number of researchers to reinforce e.g. the biochemical expertise of the team. Collaboration with cryoEM experts and with pathogenesis experts would be an added-value to the success of the projects.

Intitulé UR / équipe	C1	C2	C3	C4	Note globale
UPR9027- LISM - LABORATOIRE D'INGÉNIERIE DES SYSTÈMES MACROMOLÉCULAIRES	**A**	**A**	**A**	**A**	**A**
PROTEIN COMPLEXES AND BACTERIAL METABOLISM [STURGIS-BOUVERET]	A	A	Non noté	A+	A
ASSEMBLY OF TRANS-ENVELOPE COMPLEXES [STURGIS-CASCALES]	Non noté	A	Non noté	A+	A
SENSING THE ENVIRONMENT AND COMMUNITY LIFESTYLE IN PSEUDOMONAS AERUGINOSA [STURGIS-DE BENTZMANN]	A	A	Non noté	A	A
MACROMOLECULAR TRANSPORT THROUGH THE BACTERIAL CELL ENVELOPE [STURGIS-LLOUBES]	A	A	Non noté	A	A
DYNAMICS AND ASSEMBLY OF MEMBRANE PROTEINS [STURGIS-STURGIS]	A	A	Non noté	A	A
SECRETION SYSTEMS AND PATHOGENICITY IN PSEUDOMONAS AERUGINOSA [STURGIS-VOULHOUX]	A	A	Non noté	B	A

C1 Qualité scientifique et production

C2 Rayonnement et attractivité, intégration dans l'environnement

C3 Gouvernance et vie du laboratoire

C4 Stratégie et projet scientifique

Statistiques de notes globales par domaines scientifiques

(État au 06/05/2011)

Sciences du Vivant et Environnement

Note globale	SVE1_LS1_LS2	SVE1_LS3	SVE1_LS4	SVE1_LS5	SVE1_LS6	SVE1_LS7	SVE2 _LS3 *	SVE2_LS8 *	SVE2_LS9 *	Total
A+	7	3	1	4	7	6		2		30
A	27	1	13	20	21	26	2	12	23	145
B	6	1	6	2	8	23	3	3	6	58
C	1					4				5
Non noté	1									1
Total	**42**	**5**	**20**	**26**	**36**	**59**	**5**	**17**	**29**	**239**
A+	16,7%	60,0%	5,0%	15,4%	19,4%	10,2%		11,8%		12,6%
A	64,3%	20,0%	65,0%	76,9%	58,3%	44,1%	40,0%	70,6%	79,3%	60,7%
B	14,3%	20,0%	30,0%	7,7%	22,2%	39,0%	60,0%	17,6%	20,7%	24,3%
C	2,4%					6,8%				2,1%
Non noté	2,4%									0,4%
Total	100,0%	100,0%	100,0%	100,0%	100,0%	100,0%	100,0%	100,0%	100,0%	100,0%

*** les résultats SVE2 ne sont pas définitifs au 06/05/2011.**

Intitulés des domaines scientifiques

Sciences du Vivant et Environnement

• SVE1 Biologie, santé
SVE1_LS1 Biologie moléculaire, Biologie structurale, Biochimie
SVE1_LS2 Génétique, Génomique, Bioinformatique, Biologie des systèmes
SVE1_LS3 Biologie cellulaire, Biologie du développement animal
SVE1_LS4 Physiologie, Physiopathologie, Endocrinologie
SVE1_LS5 Neurosciences
SVE1_LS6 Immunologie, Infectiologie
SVE1_LS7 Recherche clinique, Santé publique
• SVE2 Ecologie, environnement
SVE2_LS8 Evolution, Ecologie, Biologie de l'environnement
SVE2_LS9 Sciences et technologies du vivant, Biotechnologie
SVE2_LS3 Biologie cellulaire, Biologie du développement végétal

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James STURGIS
Directeur

Marseille 18th April 2011

Reply to the AERES report

First of all, and on behalf of the members of the Laboratory, I would like to take this opportunity to thank whole-heartedly the members of the committee for their work in producing a constructive and thorough report. I find, in general, the report is a fair reflection of the laboratory, shows much good sense in the advice and opinions given, and nowhere are the opinions unreasonable or incomprehensible to me.

Second, I would like to deplore the fact that I received the report only a few days before the meeting at which the AERES committee presidents decide on the final grading of the units, and research groups. As a result I did not have the opportunity to write this letter before the meeting and thus correct any errors in the report, and bring the points raised below to the attention of the AERES at an appropriate time. I find this situation even more unreasonable since other units were given this opportunity.

Below I present several comments on the report, and then two of the research group leaders have comments

General Comments

Many of the comments in the report are in agreement with our own evaluation and this is comforting and encourages us to implement some of the recommendations rapidly. Especially concerning management objectives and the life of the research unit. In particular we are readily receptive to the comments on: the frequency of management meetings, the creation of a Journal Club, the fostering of internal collaborations and applications for European financing.

I would like to return to one of the criticisms raised in the report concerning the directors scientific vision for the development of the unit. I feel that this comment is perhaps in part due to a certain pragmatism concerning the role and power of the unit director on my part, and on the part of the foreign members of the committee a lack of understanding of the role of the research unit. I would like to take this opportunity to assert that I have a clear vision of where I think the unit should be heading and how it might arrive there, I am strongly involved in the current re-organisation of biological sciences in Marseille associated with the fusion of the three universities and am particularly vigilant on all that concerns the role and position of the laboratory and the institute IMM of which it is part. However, in the current context, both local and national, a unit director has little latitude to implement a proactive development strategy.

Comments team 1

Team 1 would like to thank you for your evaluation. We have carefully read your report on our research

The report mentions (p.8 §3) that "out of a total of 23 papers, group members have signed in first or last position in only 8 of these". In fact, our group has published in first or last position 12 papers and the group leader is co-corresponding author on a further 2 (N°1 and 4). Moreover, among the 9 remaining articles, 2 were corresponded by the previous group leader while he was still a member of the group (2006 - 2007). Therefore only 7 papers among the 23 are true collaborative papers corresponded by collaborators, thus indicating that most of our manuscripts are under our own authorship. Another consequence of this rectification means that the number of papers published in first or last position from a member of the group is now: 4 J Bact, 2 JBC, 1 Env Microbio, 2 Microbiology, 1 Appl. Env. Microbiol, 1 Int. J.Med. Microbiol., 1 Int. Microbiol.

The report mentions (p8 §2) that "the exact output of the former group leader cannot be fully assessed". We would like to emphasize the fact that since the departure of the previous group leader (January 2008), he is not a corresponding author on our 6 last research papers (N°1, 3, 4, 7, 8, 9). We have also independently obtained 3 research grants (ERAnet, FRM and VLM).

The report mentions (p8 §7) that "the team leader has been invited for 1 talk". In fact, the team leader has been invited to 3 international meetings (ASM conferences in Crete in 2006 and San Diego in 2010, IUMS conference in Istanbul in 2008) and 5 national and international departmental seminars (Pasteur Institute in 2008, SANOFI in 2009 and in 2010, Wisconsin University in 2009 and CEA Cadarache in 2010).

In conclusion the report mentions several times that the major concern of our group is the lack of focus and the too large number of collaborations in which we might not have the lead. Even though this statement should be partly reconsidered in view of the new balance in the leadership on our papers (14 over 23 instead of 8 over 23), we agree on these two points. We will therefore concentrate on the most promising topics in order maintain an international leadership. We moreover appreciate that the committee recognize the high motivation and ambition of our young team as well as its attractiveness.

Comments team 3

Team 3 thanks the committee for its support. Concerning the important downsizing that a post-doc with considerable experience of *Vibrio cholerae*, has been recruited for two years on the CTX project, and we have contacted members of the IMR laboratory on the campus to initiate nmr studies of the CTX phage capsid protein with Tol and pilus proteins. As indicated in the recommendations we would like to point out that new investigations of structure function relationships in membrane proteins are under way: using epr spectroscopy, in collaboration with the BIP laboratory on campus; using electron microscopy, and using analytical ultra-centrifugation in collaboration with the IBS in Grenoble.