Private & confidential: *Please be aware that the contents of this form may be made public resulting from the "Freedom of information Act". Personal details will not be released.*

G: NON-TECHNICAL SUMMARY (NTS)

NOTE: The Secretary of State considers the provision of a non-technical summary (NTS) is an essential step towards greater openness and requires one to be provided as part of the licence application in every case. You should explain your proposed project clearly using non-technical terms which will be understandable to a lay reader. You should avoid confidential material or anything that would identify you, or others, or your place of work. Failure to address all aspects of the non-technical summary may render your application incomplete and lead to it being returned.

This summary will be published (examples of other summaries can be viewed on the Home Office website at http://scienceandresearch.homeoffice.gov.uk/animal-research/).

(WORD LIMIT: 1000 WORDS)

Please complete the following:

<table>
<tr><td>Project Title (max. 50 characters)</td><td colspan="3">Skin homeostasis and wound healing</td></tr>
<tr><td>Key Words (max. 5 words)</td><td colspan="3">Chronic wound healing, infection.</td></tr>
<tr><td>Expected duration of the project (yrs)</td><td colspan="3">5 years</td></tr>
<tr><td>Purpose of the project (as in section 5C(3)[1]</td><td colspan="3"><table>
<tr><td>Basic research</td><td>Yes</td><td></td></tr>
<tr><td>Translational and applied research</td><td>Yes</td><td></td></tr>
<tr><td>Regulatory use and routine production</td><td></td><td>No</td></tr>
<tr><td>Protection of the natural environment in the interests of the health or welfare of humans or animals</td><td></td><td>No</td></tr>
<tr><td>Preservation of species</td><td></td><td>No</td></tr>
<tr><td>Higher education or training</td><td></td><td>No</td></tr>
<tr><td>Forensic enquiries</td><td></td><td>No</td></tr>
<tr><td>Maintenance of colonies of genetically altered animals[2]</td><td></td><td>No</td></tr>
</table></td></tr>
</table>

[1] Delete Yes or No as appropriate.
[2] At least one additional purpose must be selected with this option.

Describe the objectives of the project (e.g. the scientific unknowns or scientific/clinical needs being addressed)	Wound healing involves a series of overlapping, highly organised processes to ensure that the skin heals successfully. When these processes are disrupted, healing is delayed. If this delay is sufficiently severe then it leads to a chronic wound. Chronic wounds are a significant global problem. The incidence of chronic wounds is currently rising because those populations most susceptible, the elderly and diabetic, are rapidly expanding. This puts increasing financial strain on the world's health services. In 2014, the annual NHS spend on wound care was over £2 billion, while in the U.S. an estimated US$25 billion is spent on their treatment. Despite the global economic and social impact of chronic wounds our understanding of delayed healing remains poor. This is particularly true for wound infection. As many as 85% of lower limb amputations are preceded by an infected chronic wound. The high amputation rate highlights the failure of current therapies to treat infected wounds. This failure is partly due to: 1. The increase in antibiotic resistant bacteria. 2. The bacteria protect themselves by forming a biofilm. Biofilms form when bacteria group together, attach to the wound bed and secrete substances to form a protective coat. Biofilm avoid the body's immune response and are resistant to antibiotics. Clinicial evidence suggest that biofilms are a key reason why some chronic wounds do not heal. However, our understanding of how biofilms delay healing and which bacterial species are the most detrimental to healing remains poor. In addition, the effects of therapies to promote healing (such as antimicrobials) on normal intact skin or uninfected wounds are often unknown. A greater understanding on which processes of wound repair therapies effect is needed to guide clinical practice. Our objectives are: 1. Develop new clinically relevant models of delayed healing (such as a biofilm infected wound model). These models will provide an essential tool to address objectives 2 and 3. 2. Study the cellular and molecular mechanisms that cause delayed healing. 3. To understand how current and new wound healing therapies affect different aspects of wound repair and normal intact skin.

What are the potential benefits likely to derive from this project (how science could be advanced or humans or animals could benefit from the project)?	The results of this project are intended to: 1. Develop and improve models of poor healing. 2. Increase scientific knowledge of the mechanisms which contribute to poor healing. 3. To improve treatment regimens by better understanding how treatments affect healing.
What species and approximate numbers of animals do you expect to use over what period of time?	3000 mice for 5 years.
In the context of what you propose to do to the animals, what are the expected adverse effects and the likely/expected level of severity? What will happen to the animals at the end?	This study is designed to better understand delayed wound healing. All wounds will be made under anaesthetic and with post-operative pain relief and monitoring. Anaesthetised mice (wild-type, ovariectomised and diabetic) will receive small (10 mm incision or 6 mm diameter excision or less) wounds on their skin so that we can understand the processes involved in wound healing. After surgery, mice will be provided with pain relief and monitored closely for any signs of distress. Distress in mice after this type of surgery is very rare; however, if there is any indication of suffering we will seek veterinary advice. In some cases bacteria/fungi will be applied to the wound to represent wound infection. Wound infection is not expected to become systemic but is likely to delay healing. Animals will be monitored closely for illness and advice sought where necessary. We will apply different factors to the wounds that we believe will enhance healing, including antimicrobial agents, in order to understand how these treatments affect different aspects of repair. Animals under any procedure will be monitored daily and appropriate action will be taken for any animal presenting with any obvious stress or discomfort. If there is any indication of suffering animals will be culled. All animals will be culled by a schedule 1 method at the end. The expected severity levels: • Application of therapies on intact skin: Mild • Wounding procedures and ovariectomy: Moderate.
Application of the 3Rs	
1. Replacement	We have expertise in the use of sophisticated cell culture (*in vitro)* and whole pig skin culture *(ex vivo)* models, which we

State why you need to use animals and why you cannot use non-animal alternatives	already use to examine skin biology and wound healing. However, these models are not able to fully replicate the complex interactions *in vivo*, such as the inflammatory response. Thus, while we cannot eliminate the requirement for animal use, the *in vitro* and *ex vivo* models will be used to guide development of animal models and to understand which processes of repair are affected by wound therapies.
2. Reduction Explain how you will assure the use of minimum numbers of animals	We will use cell culture (*in vitro)* and whole pig skin culture *(ex vivo)* assays where possible to examine individual processes relevant to wound repair and will use these prior to *in vivo* work. To plan for our animal work, we have consulted a statistician to establish the minimum number of animals required for each study. Also, where possible, we will use two wounds per animal to reduce the number of animals required.
3. Refinement Explain the choice of species and why the animal model(s) you will use are the most refined, having regard to the objectives. Explain the general measures you will take to minimise welfare costs (harms) to the animals.	Mice are the most widely used species for skin research. We have many years of experience in this field and hence models have already been extensively optimised. For example, we have shown that hair cycle influences the rate of wound healing; in all studies we now assess the stage of the mouse hair cycle prior to wounding and only wound animals when hairs are in a specific hair cycle stage. By performing this step we reduce variability and group sizes within experiments. In addition, we have statistically analysed the variation in healing between two wounds on the same mouse compared to wounds from different mice. This has allowed us to refine our experiments to use a wound (not animal) as a biological replicate, therefore reducing the number of animals required. We are also aware that the time of day when mice are wounded may affect healing; therefore we will plan to perform all studies at the same time of day to minimise variation in data.
For Office Use Only	
Will the project be subject to Retrospective Assessment?[1]	Yes | No | Date due[3]:

[3] The retrospective assessment should be completed, agreed with the establishment AWERB, and submitted to the Home Office within 3 months of this date (or when the project terminates if earlier).