# A review on the issues and concerns raised in the report, “The Ugly Truth – a BUAV investigation at Wickham Laboratories”.

## ANIMALS SCIENTIFIC PROCEDURES INSPECTORATE

November 2010

# Wickham Laboratories:
# An Animals Scientific Procedures Inspectorate Report - 2009/10

(Numbers in brackets indicate page numbers)

# 1. Executive Summary

## *a. Key Findings*

Some of the allegations made by the British Union for the Abolition of Vivisection (BUAV) against Wickham Laboratories have been substantiated. Several other allegations have not been proven and some allegations arose through either misunderstanding or mis-interpretation.

Where there is *prima facie* evidence of an infringement further action will be pursued separately on the relevant Licence and Certificate files.

A. In respect of the allegation that the licensing of the mouse bioassay median lethal dose (LD50) test for routine botulinum toxin batch testing should not have been allowed – for which the BUAV believes there are existing more humane alternatives available; this allegation has not been substantiated. The authorised end point to alleviate suffering in these assays is observation and appropriate intervention by humane killing. Paragraphs 38 to 47 of the Report summarise the basis for granting the Licence.

    (i) In respect of the allegation that Wickham Laboratories are in routine breach of section 10(2A) of, and schedule 2A to, ASPA by failing to give analgesia, this allegation has not been substantiated.

    (ii) Botulism (the clinical manifestation of intoxication by botulinum toxin) is a condition producing flaccid paralysis of muscles. It does not cause muscle spasm or other directly painful effects. Indeed, botulinum containing products are used medicinally to relive various types of pain; both associated with muscle spasm and as a therapy for migraine.

    Distress associated with respiratory paralysis is not alleviated by use of analgesics – although respiratory depression, further adding to the respiratory distress will occur following administration of some analgesics.

    The administration of analgesics would not alleviate the harm to the mice and might adversely affect the residual respiratory function in such animals

B. Similar concerns were alleged about the licensing of rabbit pyrogenicity and abnormal toxicity tests; these allegations have not been substantiated. (Paragraphs 48-71)

C. Allegations were made as to how the specified humane endpoints for the mouse bioassay were applied in practice, and in particular Wickham's staff instruction on when to apply them; the consequences of this was that routinely mice were found to have died *in extremis* rather than to be have been euthanased at an earlier and more appropriate end point. It is believed this may

have caused additional unnecessary suffering to protected animals in contravention of Condition 6 of the Project Licence. This allegation has been substantiated with respect to the wide range of intervals over which clinical observations were made leading to the inconsistent application of humane end points. (Paragraphs 74–96)

D. Concerns were expressed regarding the incompetent application of humane killing methods to mice leading to unnecessary suffering; this allegation has been substantiated with respect to poor practices for both killing by cervical dislocation and by use of exposure to rising concentrations of carbon dioxide. (Paragraphs 129–140)

E. Allegations were made as to the accuracy of recent answers to written Parliamentary Questions on the licensing of death as an endpoint in toxicity testing; this has not been substantiated. Whilst it is accepted that animals will suffer adverse effects during application of regulated procedures of substantial severity, the authorised end point is appropriate observation and intervention by humane euthanasia.

F. Concerns were expressed that the botulinum products tested by Wickham may be used off-label for cosmetic purposes (in conflict with stated policy not to allow the testing of cosmetic products and ingredients); these allegations comprise two issues; that of testing a cosmetic product/ingredient and that of the cosmetic use of a medicinal product. There is no evidence that there has been testing of cosmetic products or ingredients. All testing of botulinum containing products undertaken at Wickham Laboratories is in support of medicinal products authorised by the UK's National Competent Authority. (The Medicines & Healthcare Regulatory Agency - MHRA). (Paragraphs 28-37)

   It is nevertheless recognised that off-label use of a duly authorised medicine is permitted under EU and UK legislation. Off-label use of botulinum containing products for a cosmetic purpose lies outside the jurisdiction of the Home Office.

G. Wickham staff attitudes towards animals and their welfare were alleged to be poor; this review confirmed that staff work in a busy environment in facilities which at times are under pressure of space. Whilst individual staff may speak casually during day to day work, and further improvements are possible in environmental enrichment the prevailing attitude of staff is one of appropriate care.

H. Concerns were expressed over the adequacy of Home Office inspections; the oversight by the Home Office of Wickham Laboratories extended to 25 visits of inspection over a five year period, coupled with assessment of applications for Personal and Project Licences during that time. This level of oversight is considered appropriate for an Establishment of this size. (Paragraphs 97–105)

I. Inspectors have identified a number of areas for improvement at this Establishment. These include with respect to the mouse bioassay the timing

and conduct of clinical observations; progress in the validation of an alternative assay for the product under test; study management to minimise possible operator bias. With respect to rabbit pyrogen testing; use of improved techniques during intravenous injections; introduction of a regime for increased exercise provisions and the need for a sustained programme of improvement in the fabric of the facilities. Not all issues raised by Inspectors have been followed through to completion by the duty holders at Wickham. (Paragraphs 106 -128)

J. Allegations were made that the position, performance and independence of the Named Veterinary Surgeon (NVS) at Wickham, a major shareholder in the company and, as managing director of the company, the employer of the certificate of designation holder was untenable; these allegations have in part been substantiated with respect to the incomplete performance of the duties expected of a NVS. Allegations as to a perceived conflict of interest among these roles are recommended for further examination. (Paragraphs 72–73, 141–164)

K. Allegations were made that inexperienced staff were practising injections on live mice; this allegation was not substantiated, although improvements in the training of new staff should be made by including a training session using mouse cadavers prior to use of injections in actual studies.

L. Comments were made on the quality of housing and access to water during some studies. All housing and environmental conditions reviewed during the period were in compliance with accepted codes of practice. Further improvements could be made in the provision of alternative sources of water during mouse bioassay studies

***General Findings***

The licence authorities granted to individuals at Wickham Laboratories under the Animals (Scientific Procedures) Act, 1986 (ASPA) were legitimately requested and appropriately assessed.

Some of the requirements and conditions placed on the Licences and Certificate of Designation were applied inconsistently and were breached (see below).

Not all individuals acting as Designated Persons under ASPA fully discharged their responsibilities at all times. This has led to differing local interpretations of acceptable practices in, for example, the application of humane end points in some studies.

For both individual animals, and where required for groups of mice, killing by Schedule 1 methods was poorly performed. Improvements in some aspects of this work at Wickham Laboratories have already been made.

***Findings with respect to Wickham Laboratories:***

- There is evidence of inconsistent application of humane end points in mouse potency bioassays. This represents a breach of Condition 6 of the relevant Project Licence. It is recommended that further action on this infringement be pursued separately against the Project Licence Holder. The range of intervals between clinical observations made on animals in these studies widely exceeded the intended one hourly interval, making it less likely that earlier intervention and humane killing would be possible. Furthermore, decisions on the timing of the application of humane end points were taken by staff which on occasion included relatively inexperienced technicians, albeit ones who had been signed off as having completed the basic in-house training in these observations.

- There is evidence of poor practice in the conduct of Schedule 1 killings of mice. This includes occasions when single animals were to be killed or when larger numbers were euthanased in a carbon dioxide (CO2) chamber. This represents breaches of Condition 17 of the Certificate of Designation and of Condition 16 of the relevant Project Licence. It is recommended that further action on these infringements be pursued separately against the Project Licence Holder and the Holder of the Certificate of Designation.

- There is cause for concern over perceived conflicts of interest in the roles of Named Veterinary Surgeon (NVS), Managing Director, majority share owner and the reporting manager for the Holder of the Certificate of Designation all being held by one individual. This finding was also raised during a previous review by the Home Office of work at Wickham Laboratories in 1992.

  The NVS or in his absence his deputy undertakes a weekly review of the clinical observations noted and recorded by Licensees/Technicians for the rabbits. Any animal which has been noted to show any clinical cause for concern, e.g. bruising, swellings, inappetance, is presented individually for examination. There is no examination by the NVS of individual rabbits prior to them being signed-on as suitable for re-use in the rabbit pyrogen tests.

  This system of work is a further example of the NVS appearing to incompletely discharge his expected responsibilities in full. This Report notes these failings and makes a specific Recommendation for the Certificate Holder to remedy any deficiency

- In his role as NVS, not all the expected functions are being carried out by the current incumbent. In particular there is no lead offered to ensure consistent application of humane end points; there is limited provision of advice on scientific or welfare matters; there is minimal involvement in the training or supervision of staff; and there is no involvement in the advice for, or conduct of, Schedule 1 killing of animals. This may represent a breach of Condition 9(a) of the Certificate of Designation. It is recommended that further action on this infringement be pursued separately against the Holder of the Certificate of Designation.

- In her role as Named Animal Care & Welfare Officer (NACWO), the post-holder incompletely fulfilled her duties with respect to monitoring of the conduct of Schedule 1 killing and should have provided more guidance on the need for stricter adherence to the Project Licence requirements with respect to the intervals between observations and the application of early humane end points. This directly impacts upon the care and welfare of the animals at the Establishment – a duty for which the NACWO bears a significant degree of responsibility.

- Training of new staff may be too hurried and appears directed to achieving a 'sign off' for Good Laboratory Practice (GLP) requirements

***Findings with respect to the Home Office:***

- The use of the European Union (EU) definition of a cosmetic, as applied by the Home Office in assessment of Licence Applications has been consistent with respect to authorising the testing of medicinal products at Wickham Laboratories. The descriptive term 'cosmetic' is used by other UK Government Departments in describing the purpose of some procedures undertaken with medicinal products.

- Home Office Inspectors have maintained a regular programme of inspections and raised issues of compliance and best practice with staff at the Establishment in the areas of study design, application of humane end points, environmental issues and development of a valid alternative to the mouse bioassay. However, when this Establishment transferred between Inspectors, the handover process varied and follow up was not always documented as being completed on issues raised by the outgoing Inspector**.**

- The Home Office should review current guidance and give further consideration to situations from which conflicts of interest may be perceived to arise and should publish any revision to their guidance in this area.

- With respect to this particular Establishment, several areas may be suggested for further consideration by the Home Office Inspectorate:

    a. More targeted review of Visit Reports from this DUE so that where recommended, follow up actions can be appropriately taken;
    b. More frequent review of the risk profile offered by the DUE and adjustment of the Inspection pattern/content to more adequately reflect this and to permit Inspection of critical phases of studies;
    c. When DUEs are assigned to another HOI, the provision of previous Visit Reports and discussion of outstanding actions for follow up are a high priority. It was noted that joint handover visits were not always completed at each period of change at Wickham Laboratories

***Findings with respect to other areas of responsibility:***

- Those agencies designated as the National Competent Authorities for granting Marketing Authorisations (or equivalents) may not always feel able to exert appropriate regulatory pressure on applicants to move away from animal based tests when a non-sentient test may be available/capable of development, validation and acceptance. Efforts should be made to reach a practical way forward so that refinements and replacements of animal based assays can be achieved.

- The MHRA accept the categorisation of botulinum containing products as medicines even when they are widely employed off-label for cosmetic uses. This may be perceived by others as 'licensing a cosmetic product'.

## *b. Summary of recommendations*

Recommendations 2, 3, 4, and 6 concern issues which may represent grounds for formal infringement actions to be taken against the relevant holder of the authorities under the 1986 Act.

In the light of formal actions taken earlier (1992/93) against this Designated Establishment, including some of the same individuals, consideration should be given to the appropriate level of any sanctions now imposed under the Animals (Scientific Procedures) Act, 1986.

***Recommendation 1***

That the Home Office and other Government Departments consider, perhaps through the Inter-Departmental Group on the 3R's, more clarity in the differing uses of the word cosmetic, so that its use as defined under EU Directive is clearly separated from its use as a descriptor term for certain types of medicinal products.

***Recommendation 2***

That the Holder of the Certificate of Designation at Wickham Laboratories be required to demonstrate to the Secretary of State that the full range of responsibilities expected of the Named Persons nominated under his Certificate of Designation are being discharged. In particular the Certificate Holder must assure the Secretary of State that the duties required of his Named Veterinary Surgeon (as laid out in the Guidance on the Operation of the Act and by Annex K – NVS Guidance - of The Royal College of Veterinary Surgeons Guide to Professional Conduct) are being met. Formal infringement action should be considered with respect to lack of compliance with Conditions 9A and 17 of the Certificate of Designation.

***Recommendation 3***

That the Holder of the Certificate of Designation at Wickham Laboratories be required to ensure, and to provide evidence of, regular and effective liaison between

those with responsibilities under the Act and with others who have responsibility for the welfare of the protected animals kept there.

***Recommendation 4***

That the Holder of Project Licence PPL 70/6936 *"Regulatory Testing of Biological Toxins"* write to explain how he will improve his supervision of those conducting work on this Project Licence to ensure that early and humane end points are consistently applied at all times. Formal infringement action should be considered with respect to lack of compliance with Condition 6 of his Licence

***Recommendation 5***

That ASPI management should review the oversight of this Designated Establishment against the existing guidance to Inspectors, including guidance on handover of Establishments to ensure that issues are now being appropriately followed through.

***Recommendation 6***

That a comprehensive review of Schedule 1 killing be undertaken by the Holder of the Certificate of Designation in order to achieve consistent improvement in both the processes and the outcomes. This review to include:

a. All staff currently registered as trained and competent in Schedule 1 killing to be assessed and where necessary re-trained, using accredited trainers;

b. Stopping the use of corridor floors for the conduct of Schedule 1 killings and provision of appropriate work-stations at normal working height for individual animal kills;

c. Revision of the Standard operating Procedure (SOP) for use of the CO2 chamber so that all mice should be able to move and change posture within a CO2 chamber when the chamber is filled;

d. Ensuring the establishment is adequately equipped to euthanase animals efficiently and humanely using carbon dioxide;

e. Revision to the SOPs and improvement of supervision such that all steps during Schedule 1 killing – including positive confirmation of death as required by the Act – are fully and consistently completed

The conduct and outcomes from this review must be to the satisfaction of the Secretary of State and completed without delay.

***Recommendation 7***

That the Home Office should give further consideration to situations from which conflicts of interest may be perceived to arise and should publish any revision to their policy and guidance in this area.

## 2. Scope of the Report

1 The Home Office Parliamentary Under-Secretary of State, Meg Hillier MP, requested on 9th November 2009 that a review be undertaken by the Animals Scientific Procedures Inspectorate (ASPI) into issues arising from a recent (November 2009)BUAV report, The Ugly Truth, in which the BUAV set out its concerns relating to animal care and use at Wickham Laboratories. The Home Office regulation and oversight of animal care and use at Wickham were also raised as concerns by BUAV.

2. A Superintending Inspector from ASPI was appointed to lead the Inspectorate review of matters arising from the BUAV Report.

3. In addition to consideration of the material set out in the BUAV Report this review included interviewing staff and reviewing documentation at Wickham, evaluating other material obtained by BUAV but not contained in its Report, and establishing the views of regulators and others about current regulatory requirements and the status of relevant alternative tests.

4. This report to Home Office Ministers sets out the findings and advice on possible actions (with respect to Wickham, the Home Office, or elsewhere). It also comments on lessons learned that might improve the regulatory system and compliance with the spirit and letter of the regulatory requirements.

5. In requesting the review, the Minister asked that two independent persons be appointed to quality assure the Inspectorate review and report writing processes – in particular with respect to, but not restricted to, issues that relate to animal welfare, licensing decisions and oversight, and animal use to satisfy regulatory requirements.

## 3. Allegations

### *a. BUAV report*

6. BUAV is a campaigning organisation committed to bringing about the end of experimental uses of animals. A report based on covert investigations undertaken by BUAV was forwarded in early November 2009 to the Home Office Parliamentary Under-Secretary of State.

7. The BUAV report - "The Ugly Truth" – on Wickham Laboratories, a contract research laboratory designated as a user establishment under the Animals (Scientific Procedures) Act 1986, - is based on the findings of a BUAV undercover investigator employed as an animal technician at Wickham for ten months from the beginning of 2009. The report was the subject of an article published in the Sunday Times newspaper on 1st November 2009.

8. The main issues and concerns raised in the BUAV report were:
    a. the licensing of the mouse bioassay LD50 test for routine botulinum toxin batch testing – for which the BUAV believes there are existing more humane alternatives available;
    b. similar concerns about the licensing of rabbit pyrogenicity and abnormal toxicity tests;
    c. how the specified humane endpoints for the mouse bioassay were applied in practice and in particular Wickham's staff instruction on when to apply them;
    d. concerns regarding the incompetent application of humane killing methods to mice leading to unnecessary suffering;
    e. the accuracy of recent answers to written Parliamentary Questions on the licensing of death as an endpoint in toxicity testing;
    f. concerns that the botulinum products tested by Wickham may be used off-label for cosmetic purposes (in conflict with stated policy not to allow the testing of cosmetics products and ingredients);
    g. Wickham staff attitudes towards animals and their welfare;
    h. the adequacy of Home Office inspections;
    i. the position, performance and independence of the Named Veterinary Surgeon at Wickham, a major shareholder in the company and, as managing director of the company, the employer of the certificate of designation holder;
    j. concerns that inexperienced staff were practising injections on live mice;
    k. and comments on the quality of housing and access to water during some studies.

9. The BUAV report concluded by calling for:
    a. the immediate withdrawal of Wickham's certificate of designation and the removal of the Named Veterinary Surgeon;
    b. an independent investigation into the extent to which the Home Office properly licences and inspects the facility, including the application of the 3Rs principle and minimising of suffering; and

c. the withdrawal of all project licences for botulinum potency tests, abnormal toxicity tests and pyrogenicity tests in other UK establishments

## *b. Background to the allegations*

10. The BUAV undercover member of staff was an Animal Technician at Wickham. Although not a holder of any authorities under ASPA, she had completed (August 2009) the required training in Modules 1, 2 and 3 for mouse, rat, guinea-pig and rabbit in preparation for an Application for a Personal Licence under the 1986 Act.

11. A Training Certificate to confirm the attendance of the investigator at a Modular Training Course has been issued by the relevant and accredited trainers (Certificate number BIO/09/403 was issued in the name of the undercover staff member on 25th August 2009).

12. Employed since 29th January 2009, she phoned in sick on 29th October and her keys were returned by the BUAV on 1st November 2009. Relevant background includes that:
    a. Modular training (in support of a proposed application for a Personal Licence under ASPA)was conducted off-site towards the end of her employment at Wickham Laboratories

    b. She was recruited from a staffing agency for whom she had worked on contract at another Designated Establishment

13. Whilst at Wickham the undercover staff member had access to extensive in-house documentation. Although such documents are kept in a room (locked at night time only), she requested access to documents, e.g. Project Licences once she was undertaking Modular training. Documents known to have been copied by video imaging include:

    a. Personal and Project Licenses
    b. Client listings, including details of compounds tested at Wickham
    c. Study data, including raw data, study reports and file notes
    d. Correspondence with clients
    e. Health records of animals at the establishment
    f. Signing in/out books for NVS and HO Inspector

In addition to normal daily hours, the Establishment requires staff to work throughout the night on occasions (to facilitate animal observations and interventions during studies). She was on rota for undertaking such duties, usually being one of two staff members on site at night.

14. She had been trained to the Establishment's regulatory procedures which cover requirements for GMP work and GLP studies and was signed off as competent to undertake the killing of mice by two Schedule 1 methods:

a. Cervical dislocation
b. Euthanasia by exposure to carbon dioxide gas in a rising concentration within a specifically designated chamber

15. In-house training records maintained at the Establishment show that the NACWO or a Senior Animal Technician trained, assessed and signed off all new staff members prior to assignment to work in the animal facilities. This training was prior to, and separate from, the Modular Training referred to above.

16. The BUAV investigator has provided a verbal summary of her involvement at Wickham Laboratories:
    a. Employment commenced 29th January 2009, work restricted to cleaning and general duties, with no animal handling required.
    b. Progressing through supervised feeding/watering of animals, she stated that some aspects of the basic training were over detailed and some were too rushed. Her Supervisor had been on leave for some of the training period
    c. Examples of areas considered by her to be too rushed included:
        i. Being placed on the night duty rota before being signed of for all the work such a job might entail
        ii. Being expected to be the 'senior' one of the two night duty staff when working with a newer staff member
    d. Training with a Senior Animal technician included clinical scoring and being shown Schedule 1 killing by cervical dislocation. Having seen the Senior Technician do it (on the corridor floor) she was then expected to undertake Schedule 1 kills during the mouse potency bioassay then running. (No technique development on mice cadavers was undertaken as part of the training)
    e. She also stated that the Senior Technician had agreed that her training was being a bit rushed. The BUAV investigator was surprised that there had been no mention of checking vital signs after cervical dislocation to confirm death. (This latter being raised by the course tutor during the Modular training undertaken towards the end of her employment at Wickham)

17. Samples of video clips were provided on two occasions by BUAV, selected from the material obtained by their investigator. Those provided in the first tranche are summarised below:

    a. Undated; 'Rabbit handling'. Shows one rabbit being lifted by the scruff/skin on the back out of the pyrogen testing stocks. 15 secs duration
    b. 11 Aug 09 'Staff Schedule 1 training on live mice'. Shows very little animal content at all. Schedule 1 (Cervical dislocation) is being undertaken on the corridor floor. A voice is heard to say "I definitely broke its back I think". 51 secs duration
    c. 23 Jul 09 'Mouse breathing'. Shows one box of mice which have been apparently subject to CO2 euthanasia. One animal is shown on checking to be still breathing and is passed by the filmer to a

colleague who states that the mice were in the chamber for 10 minutes and then kills it by cervical dislocation. 57 secs duration

d. 3 Aug 09 'Killing mice'. Shows mice being killed by cervical dislocation (on a bench top) one comment is heard "That ones back went before its neck" also "That was a good one that was" A few animals showed post-procedural twitching for longer than might be anticipated. 2 mins 39 secs duration
e. 11 Jun 09 'Rabbit pyrogen injections'. Shows rabbits in purpose built stocks for the study, none are distressed. Filmer is moving equipment and 'clattering' things and is heard to be advised to hold animals in an alternate manner so as to facilitate the injection of material by the other technician (e.g. around 8 mins "Don't hold his head so tight; no, keep his butt in and across" and again around 18 mins "You may be gripping too hard – that's not too good".) The original dosing technician makes two attempts to dose one animal and then seeks help from another member of staff who successfully completes the procedure with the original dosing technician holding the rabbit. There are taped comments such as "Hamish you're a little shit" and "You're a disgrace" which are made to the animals, but on no occasion does the tone or volume of the speaker change from their normal manner. All video shots of animals in the stocks show placid animals. 43 mins 31 secs duration.

18. Those in the second set of clips are also summarised:
    a. 11 Aug 09 (1). Filmer observes that some mice appear to be on their "last legs". A second staff member says to leave them till next observation time in an hour. There are two fleeting views (less than 5 secs in total) of mice showing adverse clinical signs (depressed activity, laboured breathing) in their home cages. 1 min 43 secs duration
    b. 11 Aug 09 (2). Shows a trainee undertaking Schedule 1 killing on corridor floor; she claims to have broken the back not the neck. Checked by second staff member. A prominent part of the clip is a third staff member stating that a dead mouse has been found on the floor of a room – discussed with others and believed to have been 'dropped' before being placed in a bag as the headcounts all tally. 47 secs duration
    c. 11 Aug 09 (3). Continuation/part of video clip 2 above. 49 secs duration.
    d. 11 Sep 09 (1). Staff inserting and fixing rectal temperature probes into rabbits on test. Transient discomfort during insertion is seen.1 min 9 secs duration
    e. 11 Sep 09 (2). Further footage of insertion of rectal probes. The Technician/Licensee is shown not wearing personal protective clothing appropriately (mask pulled down leaving nose uncovered). 1 min 7 secs duration.
    f. 12 May 09 (1). Conversation over injection training and staff failing to be allowed to do any injections after being signed off. Video also shows an overcrowded CO2 chamber being used for Schedule 1 killing. 3 mins 17 secs duration.

g. 12 May 09 (2). Staff member confirms that Schedule 1 killing by cervical dislocation is not practised on cadavers before using the technique during live studies. States that she used to be "the worst back breaker in the world" at one time. 53 secs duration.
h. 12 Aug 09. A staff member is shown taking a live mouse from a cage on a workbench, Schedule 1 killing it, and returning it to the original cage in which there are live mice. 40 secs duration.
i. 15 Apr 09. Film of an overcrowded CO2 chamber for Schedule 1 killing. (Alleged to be 65 mice in it but this is not verifiable on the video). 1 min 39 secs duration
j. 23 Jul 09. One staff member is asked to 'stop banging about' in the rabbit pyrogen testing suite. Another staff member is using personal protective mask inappropriately and wearing it below her nose. 2 mins 29 secs duration
k. 25 Jun 09. Rabbit in floor pen with a radio on (speech station) in the background. 1 min 29 secs duration.

19. The BUAV have not released, despite repeated requests for such, to the Home Office the complete data, documents and material collected by them during the undercover investigation.

## 4. Inquiries Pursued

### *a. Were authorities under A(SP)A in place for the work undertaken at Wickham?*

20. There are three Project Licences (PPLs) held by persons employed at this Designated User Establishment (DUE). They are authorised for the purposes, (as defined within ASPA) of Control of Disease, Ill-health or Abnormality.

    a. **PPL70/6365** ***"Development of Alternative Assays for Biological Toxins"*** authorises a programme of work comprising two objectives within that programme of work:

        1. Development of a non lethal in-vivo assay to develop an alternative to the mouse LD50 procedure currently used for the assay of botulinum toxin in the pharmaceutical industry.

        2. Development of an in-vitro assay to identify a process leading to the development of an in-vitro alternative to the mouse LD50 procedure currently used for the assay of botulinum toxin in the pharmaceutical industry.

    ii. This work is ongoing at the DUE to replace, in part or in whole, the use of the mouse bioassay currently required by regulatory authorities for licensing and marketing of medicinal products containing botulinum toxin.

    iii. Work at this DUE is in collaboration with a UK National Institute which is referenced in the allegations from BUAV. The work is supported by one of the main commercial sponsors for the bioassays; providing reference toxin; assisting with study design and interpretation; and collaborating with both Wickham and the other Institute in dissemination of any findings.

    iv. The BUAV investigator did not participate or comment on aspects of this Licence.

    b. **PPL 70/6417** ***"Safety and Quality Control testing of Pharmaceuticals and Medicinal Devices"*** authorises a programme of work with one broad objective;

        1. The production of valid quality control data in support of pharmaceutical products and medical devices

    ii. This work includes the performance of the rabbit pyrogen test conducted when it has been shown that non-animal alternative

tests are not suitable. The PPL holder is obliged to confirm that the legislation and/or regulatory authorities will not accept *in vitro* data on pyrogenicity for the specific material to be tested.

c. **PPL 70/6936 *"Regulatory Testing of Biological Toxins"*** authorises five objectives within the programme of work:

    1. To undertake testing procedures to ensure the safety, efficacy and overall quality of biological toxins and associated proteins used for medicinal products

    2. To provide testing services to assist with product development and clinical trials associated with biological toxins and associated proteins.

    3. To provide testing services for quality control and compliance with Good Manufacturing Practice for products associated with biological toxins and associated proteins

    4. To provide testing services to assist with pharmacovigilance of drug products associated with biological toxins and associated proteins.

    5. To provide testing services to assist with health and safety of operators involved with biological toxins and associated proteins.

    ii. Work with botulinum toxin is conducted in knowledge of alternative technologies, some of which are under concurrent development at this DUE.

d. Eleven Personal Licences (PILs) are held by persons working at this establishment. All are current and are appropriately authorised for the conduct of studies on the above Project Licences. PIL Holders were observed, both by Inspectors on visits of inspection and by the Reviewing SI, undertaking injections, performing clinical observations of animals on studies and implementing Schedule 1 killing of animals. This work was conducted under what was considered by the PPL holder the appropriate level of Supervision of the PPL holder, although no evidence was found of direct supervision of the procedures.

e. The application of humane end points in mouse bioassays followed the instruction from the PPL holder as to what was the appropriate point of intervention – it was not left to an individual PIL Holder's judgement. Whilst PIL Holders did not challenge the instruction of the PPL holder, the failing is considered to be one of failure by the PPL holder to set the correct earlier intervention in mouse bioassays.

f. Application of humane killing as an appropriate earlier end point was not restricted to PIL holders; the undercover investigator undertook clinical observations and decided on the appropriate intervention – euthanasia or not – despite not having a PIL.

g. The Establishment is designated under ASPA and a holder of the Certificate of Designation (PCD) has been appointed. In turn the PCD Holder has nominated a veterinary surgeon intended to fulfil the role of Named Veterinary Surgeon (NVS) under the Act and a person to be in charge of day to day animal welfare – the Named Animal Care & Welfare Officer (NACWO). These nominations have been accepted by the Secretary of State

h. The carrying out of killing of animals by a method approved in Schedule 1 of ASPA does not require any specific authorisation under the Act.

    i. The holder of the Certificate of Designation (PCD) is required under the Act to ensure that only appropriately trained and competent persons carry out killing by methods listed in Schedule 1
    ii. A Register of those trained and competent to kill using methods listed in Schedule 1 must be maintained by the holder of the Certificate of Designation and made available to the Home Office on request
    iii. Holders of Personal Licences (PILs) are required to arrange for any animal which has reached the conclusion of procedures and is suffering or is likely to suffer adverse effects to be promptly and humanely killed.
    iv. Holders of Project Licences (PPLs) are required to ensure that animals which have reached the conclusion of regulated procedures are promptly and humanely killed by either a Schedule 1 method or by another authorised method

21. The BUAV report also raised the question of whether the Home Office should issue 'generic' licences which do not name the substances to be tested but rather grant authority for defined types or classes of materials.

    a. The 1986 Act, Section 5 (1) states: "*A project licence is a licence granted by the Secretary of State specifying a programme of work and authorising the application, as part of that programme, of specified regulated procedures to animals of specified descriptions at a specified place or specified places*".

    b. This is further clarified in section 5.3 of the Guidance on the Operation of the Act: "*A single project licence is expected to cover a coherent programme of work managed by an individual to meet a common set of well-defined objectives. A licence might therefore cover the entire medicinal drug discovery process, involving large numbers of animals of a wide range of species, numerous protocols, and a large team of*

*personal licensees. Or, in contrast, a licence might cover the work of a single scientist, involve the investigation of one part of one system, and use a small number of animals of a single species*"

c. Project Licences, where authorised, are prospective authorities in force for up to 5 years from the date of issue. Future developments in science, biomedical research and associated technologies and the obligation to take account of advances in the 3R's mean that it is not practical or necessary to list every possible unique substance which may be tested during the prospective life of each Project Licence

d. Licences will include requirements for the duty holders (PPL holders) to ensure that any regulatory requirements are current; that where non-animal alternative tests are valid and acceptable they are preferentially used, and that a justification is held on a case by case basis for the conduct of animal based testing. The phrasing of the authorities as stated above are intended to be sufficiently clear to set a framework of responsibility for such testing.

22. The Duty Holder responsible for ensuring that a particular substance proposed for testing does indeed meet the criteria specified in the Project Licence Authority is the Project Licence Holder. On challenge from the Home Office Inspector or on request from the Secretary of State, it is expected that the relevant PPL holder can substantiate why a particular material has been tested.

23. The PPL holder at Wickham Laboratories has stated that sponsors requesting testing of materials in the rabbit pyrogen studies have a specific regulatory need for their material to be tested in this manner. An addendum to each commercial contract requires the sponsor to signify the status of their request:

a. "ADDENDUM RELATING TO ANIMAL TESTING

All tests involving the use of animals are undertaken for Q.C. and Safety purposes as defined below:-

Please tick as appropriate

Pharmaceutical products – Quality Control testing as specifically required by appropriate legislation in the country of manufacturer and/or use (e.g. in the European Community 2001/83/EC and 2003/94/EC for human pharmaceuticals and 2001/82/EC and 91/412/EEC for veterinary pharmaceuticals; in the United Kingdom by the Medicines Act 1968; in the United States of America by the Food and Drug Administration).

Medical devices – Safety tests as required by the European Medical Device Directive (93/42/EEC) and in the United States of America by the Food and Drug Administration.

If the testing is required for any other purposes Wickham must be informed in advance to confirm that appropriate clearance is available to undertake the work, please give details."

24. The Establishment holds current (and prior) certification for Good Laboratory Practice (GLP), Good Manufacturing Practice (GMP) and other national or international accreditation relating to the conduct of laboratory work. They do not specifically retain in-house expertise in regulatory affairs, although GLP, GMP and ASPA responsibilities require some degree of knowledge of regulatory affairs.

    Wickham Laboratories also offer an *in vitro* assay for testing for pyrogens and has demonstrated that they have encouraged sponsors to validate a product-specific, non-rabbit alternative test based on a regulatory acceptable method.

25. For all substances except one identified by the BUAV in Appendix 3 of their report there is a client-based justification held on file by Wickham Laboratories, although some of the justifications pre-date the date of issue of the current Project Licence.

26. Since 2010 (associated with this Review of their practices) the Establishment have extended their requirement so that sponsors must submit justifications on a product by product basis.
    a. The requirement for clients to undertake the testing relates to the specifications that they have in their Manufacturing Authorization and Product Licences which may not necessarily be pharmacopeial methods
    b. The one product for which no justification is currently held on file is glucose (dextrose) monohydrate for parenteral use as a pyrogen free solution. There is a pharmacopoeial requirement that this material is demonstrably pyrogen free and a National Competent Authority may require such pyrogen testing to be undertaken in rabbits.

27. ***Conclusion***: Licence and Certificate authorities to permit work under ASPA 1986 were, and are, in place at Wickham Laboratories.

## *b. Were these authorities granted legitimately by the Home Office?*

The three Project Licences referenced above were subject to consideration and assessment by ASPI before the assigned Inspectors made advisory recommendations with respect to the granting of the Licences. During assessment the issues considered included:

a. Appropriate categorisation of materials as medicinal products v cosmetics
b. The regulatory need for testing
c. Availability and suitability of non-animal alternatives;
d. Design and conduct of proposed studies;
e. Provision for recognition and control of adverse effects on animals;
f. Appropriate severity limit for study protocols.

A recommendation from ASPI to the Secretary of State that an application should be granted a Licence implies that the conditions stated within the Licence are essential to the compliant operation of that Licence. This includes requirements for justifications on a case by case basis when the Licence authorises work under a framework as an authorised Programme of Work

**Botulinum Toxin testing**

28. The UK's National Competent Authority (NCA) with respect to licensing medicines is the MHRA (Medicines and Healthcare products Regulatory Agency), who have confirmed the Home Office Inspectorate opinion that the product Dysport (from Ipsen Biopharm) has a UK Marketing Authorisation with licensed claims for medical disorders. It does not have an authorised claim for cosmetic use.

29. The phrase 'cosmetic use' or cosmetic claim' is perceived by MHRA to be misleading as the agreed medicinal claim for the botulinum toxin containing product Dysport is for "The temporary improvement in moderate to severe glabellar lines seen at frown, in adult patients under 65 years of age, when the severity of these lines has an important psychological impact on the patient".

30. The assessment by MHRA is that although the use of injectable botulinum toxin produces a change in appearance, i.e. a cosmetic effect, it is the psychological impact which is receiving treatment. As such products are outwith the EU definition of a 'cosmetic' *(vide infra)* and as the Commission on Human Medicines (CHM) has agreed (as the Independent Advisory Committee to the MHRA) with the medical claim, MHRA are regulating the products as medicines.

31. The following definition of a cosmetic has been agreed by the EU and is used by the HO when implementing authorities under ASPA :

> "any substance or preparation intended for placing in contact with the various external parts of the human body (epidermis, hair system, nails, lips and external genital organs) or with the teeth and mucous membranes of the oral cavity with a view exclusively or principally to cleaning them, perfuming them or protecting them in order to keep them in good condition, change their appearance or correct body odours". (From Article 1(1) of Directive 76/768). http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:1976L0768:20080424:en:PDF

Under this definition, the use by injection of botulinum toxin cannot therefore be included and, as such, it would be defined under Directive 2001/83 as a medicine. http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=CONSLEG:2001L0083:20070126:EN:PDF

32. The testing of a licensed medicinal product, such as botulinum toxin products defined by the NCA as, and intended for use as, a medicine is permissible under ASPA.

33. There are differences in the meaning of 'cosmetic' which apply in differing scenarios:

    a. As defined by the EU when referring to a particular substance or preparation (see above)

    b. As a cosmetic function of a medicine which is given for an authorised medicinal claim, (e.g. botulinum toxin)

    c. As a medical device (e.g. collagen or breast implants for reconstructive and aesthetic/cosmetic surgery)

***Provision of Advice from MHRA on off-label uses***

34. The MHRA website contains explicit advice on the supply and administration of botulinum toxin products in cosmetic procedures. This advice includes by product name, Dysport – which has no 'cosmetic procedure' claim. Discussion with MHRA during this review failed to identify how this information was placed on their website, and an internal referral by MHRA to the MHRA Enforcement Unit has been made. http://www.mhra.gov.uk/Howweregulate/Medicines/Availabilityprescribingsellingandsupplyingofmedicines/Frequentlyraisedissues/BotoxVistabelDysportandotherinjectablemedicinesincosmeticprocedures/index.htm

35. It was stated that the advice from MHRA only refers to the protection of public health and as such the advice which includes 'off-label use' is intended to clarify what the legitimate channels are for the supply and administration of products.

36. The Department of Health has commissioned the Independent Healthcare Advisory Service (IHAS) to produce guidance for patients on 'Standards for Injectable Cosmetic Treatments'. This document, Published August 2008, has been referenced during the current review: http://www.independenthealthcare.org.uk/index.php?/component/option,com_phocadownload/Itemid,63/download,197/id,16/view,category/

    a. Its title refers to injectable cosmetic treatments; but as the EU definition of a cosmetic excludes injectables, the use of 'cosmetic treatment' here is taken to be as an adjective rather than as a noun.

    b. Standard 3 (3.10) of the IHAS document states "Medication shall not be used outside of the licensed indications ("off-label") where there is a product available that can be used within licensed indications. Where "Off Label" use of medicines does occur the patient's specific consent must be given. "

37. Whilst the HO has publicly stated, and confirmed, that no testing of cosmetics will be authorised in the UK, other Government Departments offer some degree of support and advice to the 'off-label' use of products described specifically as injectable cosmetic treatments. Whilst this latter use of 'cosmetic' is as a descriptor rather than as a substance, confusion is still likely to exist.

***Advice on use of Alternative Testing***

38. MHRA strongly believe that they have only a limited role in enforcing the adoption of non-animal alternatives by holders of Marketing Authorisations. They have stated that it is beyond their legal competence to compel applicants to move to alternative based tests.

39. The MHRA do however compel applicants to change submitted, validated methods in other areas of assessment if they believe the alternative method will improve the protection of human health. As an example of this they would compel a manufacturer to change from astic production (which has an inherent risk of broaching the sterility of products at several stages) to terminal sterilisation (which is more likely to ensure product sterility) if the medicinal product is stable enough to permit terminal sterilisation.

40. In cases where a pharmacopoeia includes more than one method (or refers to 'other methods if validated') applicants are at liberty to choose which method they submit for MHRA assessment. The MHRA do not seek to make changes or to encourage adoption of non-animal alternatives as they believe that they do not have the powers to do so. This policy has never been tested in court.

41. The MHRA agreed that their stated support for the 3Rs is heavily qualified by what they perceive as their first priority; the protection of public health, and feel that they could not legitimately refuse or delay a marketing authorisation on the grounds of animal welfare or use of specific animal/non-animal testing regimes.

42. In Europe, $LD_{50}$ testing of botulinum toxin, i.e. the bulk drug substance and the final drug product lot, is a requirement of the monograph 2113 included in the European Pharmacopoeia (Ph Eur) 6.0 "Botulinum Toxin Type A for Injection". This requirement has been in force since January 2005. European Pharmacopoeia monographs and texts are legally binding quality standards for all medicinal products in the EU.

43. The Ph Eur reference method for potency is the $LD_{50}$ assay. Nevertheless, the monograph allows the possibility to develop alternative methods, in particular in the interest of animal welfare and the 3Rs. Three different alternative approaches are noted in the Ph Eur but users are not limited to these methods, which are: "*an endopeptidase assay in vitro, an ex vivo assay using the mouse phrenic nerve diaphragm and a mouse bioassay using paralysis as the endpoint."* Furthermore, it is stated in the Ph Eur that alternative methods must be suitably validated on a product by product basis in comparison to the reference method, which is the $LD_{50}$ assay: "*After validation with respect to the $LD_{50}$ assay (reference method), the product may also be assayed by other methods that are preferable in terms of animal welfare, including one of the above mentioned."*

44. The HO has contributed partial funding to the development of an alternative method for the determination of potency of botulinum toxin. The view has been taken that responsibility for product specific validation and eventual acceptance of such an alternative remains firstly with the holder of the Market Authorisation and secondly with the MHRA as the relevant Competent Authority.

45. With respect to potency testing of botulinum toxin, there are several published and researched methods which can contribute information on some aspects of the toxin. A recent summary (2009) by the National Institute for Biological Standards and Control (NIBSC) researchers and published on the UKs NC3Rs (National Centre for the Replacement, Refinement and Reduction in Animal Research) website confirms the widespread regulatory requirement for mouse bioassay data, whilst also reviewing alternative strategies: http://www.nc3rs.org.uk/downloaddoc.asp?id=853&page=1023&skin=0

    i. "Although the mouse LD50 assay is recommended for use by national and international regulatory authorities as the primary assay method for use prior to marketing, a number of alternative methods and approaches which minimise the distress caused to test animals, use more humane endpoints, or which could replace their use, have been developed and adopted."

    ii. The current European Pharmacopoeia Monograph 2113 requires the use of the mouse bioassay for release of batches of product containing botulinum toxin. The monograph states that fully validated alternatives must satisfy the criteria of that Monograph in all respects.

1. A flaccid paralysis assay in mice is less severe in the effects on mice and has been validated at a few laboratories. This assay however requires as a pre-requisite the determination of potency in the mouse LD50 bioassay.
2. The synaptosomal associated protein (SNAP-25) assay determines the toxin potency of the material but does not assay the other domains of importance. (It assays only the toxin light-chain activity, and changes to other toxin domains, are not detected). This assay can
    a. Accurately quantify small (ng level) quantities of active toxin in finished product
    b. Detect toxin in presence of high concentration of bulking and stabilising material
    c. Provide high sensitivity with limit of detection equivalent to or better than mouse LD50
    d. Provide high precision and reproducibility
    e. Encourage transferability between laboratories.
    f. However, it is only relevant for testing of final lots derived from the same bulk toxin, for a particular product once mouse LD50 data has been generated as a reference potency.

3. The mouse phrenic nerve/hemi-diaphragmatic assay (HDA) is an *ex vivo* assay which requires considerable technical expertise to set up and run. Variability in the results obtained in this assay require large group sizes of preparations to be set up and again there is inconsistent correlation with the 'gold standard' of the mouse bioassay
4. The rat inter-costal neuromuscular assay could permit *ex vivo* assessment, although studies reported to date show that the variability is greater than for the HDA.
5. Cell based assays have been reported using murine neuroblastoma: Neuro-2a (ATCC) cells which are sensitive and specific for the actions of botulinum toxin type A. This method is still in development and does not as yet show sufficient sensitivity or signal-to-noise ratio in its results. This precludes its use at present for determination of final lot potency in product release assays.
   http://www.ipsen.ltd.uk/batch_potency_botulinum_10.pdf
6. Other assays such as the use of matrix assisted, laser desorption ionisation , time of flight mass spectrometry (MALDITOF) or high performance liquid chromatography, electro-spray ionisation tandem mass spectrometry (HPLC ESI MS/MS) have been demonstrated to have very low limits of detection but

have not been validated for complex biological media with respect to potency or safety.

iii. Workshops have been run in the EU (2009) (In preparation) and USA (2008) http://iccvam.niehs.nih.gov/docs/biologics-docs/BoNTwkshprept.pdf under the auspices of national and international regulatory bodies to agree improvements in the botulinum toxin testing assays. Due to the lack of valid interchange of data obtained from different products – any potency data or other assay is product specific – these meetings have concluded that the mouse bioassay remains the 'gold standard' for testing of botulinum containing products

46. ***Conclusion***: The authorities granted by the Home Office under ASPA were issued legitimately and with appropriate reference to current requirements for authorised medicinal products.

47. Whilst alternative assay methods could be developed and validated for the specific products being tested at Wickham Laboratories, this has not yet been completed. The criteria applied through the Marketing Authorisations for medicinal products allow for a manufacturer to state the pharmacopoeial method they choose to employ.

***Recommendation 1***

That the Home Office and other Government Departments consider, perhaps through the Inter-Departmental Group on the 3R's, more clarity in the differing uses of the word cosmetic, so that its use as defined under EU Directive is clearly separated from its use as a descriptor term for certain types of medicinal products.

**Pyrogen Testing**

48. The authorisation for use of an animal based pyrogen test is provided in one of the Project Licences to permit the testing of pharmaceutical and veterinary products in order to ensure that they are free from extraneous contaminants or unacceptable levels of biological activity that could cause toxicity or fever when administered to humans or animals.

49. Any testing in animals can only be conducted when the objectives cannot be achieved by non-animal methods such as chemical analysis or *in vitro* tests. Testing is conducted according to approved test methods such as those detailed in the European or British Pharmacopoeia, USP, Japanese or Russian Pharmacopoeia. The Project Licence would also permit the use of animals when this is essential for validation of alternative (non-animal) tests for the determination of abnormal toxicity and pyrogenicity.

50. Wickham Laboratories have held authority to conduct pyrogen tests in rabbits under a previous PPL and have alternatively undertaken *in vitro* testing (Limulus amoebocyte lysate assay – LAL) when appropriate. The use of rabbits is only where non-animal alternatives have been shown to be

unsuitable or where a regulatory authority requires rabbit based data to be generated.

51. *In vitro* testing may be unable to detect certain classes of pyrogens (non-endotoxin pyrogens) or may be subject to assay interference from (typically) proteins or adjuvants present in the test formulations. In such cases it is a regulatory requirement that a rabbit based pyrogen test is undertaken to assure product quality.

52. In cases where a Marketing Authorisation for a (human or veterinary) medicine was granted with the stipulation that freedom from pyrogens must be demonstrated using a rabbit pyrogen assay, the manufacturer is obliged to continue testing each batch of product – or to attempt *de novo* product-specific validation of an *in vitro* alternative. As considered above, the driver to move from rabbit based to a non-animal test is not considered to be the responsibility of the regulatory NCA, but rather that of the manufacturer.

53. Progress continues to be made in validating alternatives which may perform as comprehensively as the rabbit model, with further guidance being issued in April 2009 by the European Medicines Agency on the replacement of rabbit pyrogen testing for plasma derived medicinal products. http://www.ema.europa.eu/pdfs/human/bwp/45208107enfin.pdf

54. A typical rabbit pyrogen test authorises withdrawal of food overnight – current and recorded practice at Wickham Laboratories is for this to be undertaken by staff on night shift work. The rabbits will be without food for a variable period of typically 10 hours prior to commencement of the test. After acclimatisation nimals are restrained in purpose made stocks so that continual recordings of rectal temperatures can be made via electronic thermometers. Tests may run for 6 – 8 hours on any single occasion.

55. Rabbits are weighed before use to determine any significant weight changes. Differences in the association with feeding, age of animal and previous body condition may alter the measured bodyweights. No evidence was found for a loss of 10% bodyweight. The tolerance of 2% loss accepted as a practical limit by Wickham Laboratories for this test does not cross any Home Office threshold of concern for animal welfare. Additional observations on animal health are also undertaken (see below). Any doubts in a decision to include individual animals in a scheduled test are referred to the NVS

56. Rabbits are not re-used if the results of the test indicate a pyrogenic response, or if certain classes of compound have been tested; e.g. cytotoxic agents, some biological materials (which may produce sensitisation). The PPL authorises reuse on up to approximately 45 occasions each year, provided that the animals are healthy, the ear veins have healed and that veterinary certification has been undertaken. A mandatory six day interval between any re-use is also set in the Licence for all animals being re-used.

57. Records of rabbit health checks undertaken by the technicians and signed off by the NVS have been examined. Low (<0.5%) incidence of any bruising on

ear veins were noted. It was confirmed from study records that for the oldest rabbit held at the DUE, there were 28 occasions of re-use in 2008 and 29 occasions in the period Jan – October 2009.

58. There were no recorded instances of injuries occurring to rabbits whilst restrained in the stocks. Animals are housed singly to prevent fighting or aggressive behaviour which frequently occurs when new rabbits are introduced into older established groups. At Wickham Laboratories the colony is subject to a low but frequent turnover as animals are withdrawn from the colony and replaced by new stock

59. The single injection (via ear vein) of the test material is not intended to produce any adverse or toxic effects, and the discomfort of restraint with a rectal thermometer inserted is regarded by ASPI as a mild procedure. A normal response to ongoing or moderate stress in rabbits would be to raise metabolic rate and hence rectal temperature. This does not occur in animals which are appropriately trained to periods of restraint in purpose-built stocks. Subject to veterinary certification (by the NVS), animals which have completed pyrogen tests may be re-used in subsequent pyrogen tests.

60. Allegations of poor injection technique during intravenous administration via the marginal ear veins of rabbits were specifically examined during interview at Wickham Laboratories. Whilst video evidence does show one Licensee experiencing difficulties, the appropriate action is taken in seeking assistance from a more senior technician. Examination of rabbit health records (on which specific notation is made of any bruising/damage to ear veins) showed very low levels of bruising following intravenous dosing

61. ***Conclusion***: The conduct of rabbit pyrogen testing at Wickham Laboratories is in support of the requirements of regulatory authorities for specific medicinal products. The Establishment is well aware of, and participates in, the use of non-animal alternative testing for the detection of pyrogens. At present not all substances can be validly tested for pyrogens using *in vitro* methods.

**Abnormal Toxicity Testing**

**62.** The abnormal toxicity testing undertaken at Wickham Laboratories is similarly a requirement of regulatory authorities for either medicinal products, vaccines or medical devices. There is acceptance by the EP that such testing may be removed from some regulatory requirements for safety testing, e.g. for veterinary vaccines, provided that sufficient data on batch to batch consistency is provided to the responsible regulatory authorities.

63. The observation schedule for animals undergoing abnormal toxicity testing includes a requirement to observe all animals immediately after dosing and at least daily for the duration of the study.

64. The relevant S 19 (b) protocol of the Project Licence additionally includes obligations for extra attention: "If any animal appears hunched, thin lethargic or in poor condition it should be brought to the attention of the NACWO immediately. Any loss of appetite or water intake should be reported similarly. In the case of mice, any animal which is in obvious distress and is vocalising should be killed immediately by a schedule 1 method. Conversely, any guinea-pig that is unusually quiet should be paid particular attention."

65. These requirements are considered sufficient for an appropriate level of observation and intervention in these studies

66. The number of Abnormal Toxicity Tests conducted is reducing at Wickham Laboratories and improvements in manufacturing methods mean that there are rarely any adverse effects seen during the studies. It is not the intent of such studies to demonstrate obvious toxic effects and as such all studies can be managed within the severity limit of 'moderate'. It would be inappropriate to assign a severity limit of 'substantial' for a study which can achieve its scientific objectives at a lower level of severity

**Development of Alternatives**

67. The need for product by product validation of alternative tests (in the case of pyrogen detection) or for batch by batch consistent analytical data (in the case of abnormal toxicity) means that animal based studies continue to be a legitimate regulatory requirement.

68. The leadership for such transition, to *in vitro* alternatives, could however be more clearly communicated, e.g. consideration given to use of a specific time period for a Marketing Authorisation during which the manufacturer would be expected to have either validated an alternative or to have conclusively shown that current *in vitro* options were not scientifically valid.

69. In responding to the question as to what pressure the Home Office has put on Wickham Laboratories to discard the LD50 for botulinum bioassay, a Project Licence has been authorised for developing a product-specific alternative test (flaccid muscle paralysis); Inspectors have frequently raised questions during inspections concerning progress of work under that Licence; and the Home Office has supported an Inspector attending a European Expert Group meeting in 2009 to review progress with alternatives specifically in this area and to try to identify further alternative strategies.

70. Several HOI reports include notes that the Inspector had continued to raise in discussion the validation and use of alternative tests, whilst recognising that the criteria applied through the Marketing Authorisations for medicinal products allow for a manufacturer to state the pharmacopoeial method they choose to employ.

71. The relevant Project Licence at Wickham Laboratories includes requirements, within Section 18 of that Licence, for clients to justify their request for any

testing on animals. In practice this means confirmation that the requirements are as laid down by the relevant regulatory authority. In cases where the Establishment is asked to undertake work for which an alternative test may be possible, the Licence requires the PPL holder to encourage the client to vary their marketing authorisation in order to replace the need for animal testing. (see paragraphs 22–27.)

### *c. Were requirements, conditions and controls contained within these authorities complied with by staff at the DUE?*

72. Persons holding positions and/or authorities under the 1986 Act must adhere to the requirements of the Act, the constraints and requirements of their Licences and the Standard Conditions as published. In addition, if further Special Conditions are placed on Licences, these must also be complied with. There were no Special Conditions placed on the Project Licences covered by this review.

    i. Condition 9A of the Certificate of Designation imposes a requirement for the Certificate Holder to be responsible for the performance of named persons under the Act.

    ii. Condition 16 of the Certificate of Designation requires the certificate holder to take all reasonable steps to prevent the performance of unauthorised procedures in the establishment, and make adequate and effective provision for regular and effective liaison with and between those entrusted with responsibilities under the Act and with others who have responsibility for the welfare of the protected animals kept there

    iii. Condition 17 of the Certificate of Designation imposes responsibilities for ensuring competence in and compliance with killing of animals by methods listed in Schedule 1 to the Act

    iv. Condition 6 of each Project Licence requires for any procedure, the degree of severity imposed shall be the minimum consistent with the attainment of the objectives of the procedure, and this shall not exceed the severity limit attached to the procedure. The minimum number of animals of the lowest neurophysiological sensitivity shall be used in procedures causing the least pain, suffering, distress or lasting harm.

73. The Certificate Holder has discharged part of his responsibilities for nominating a Named Veterinary Surgeon and a Named Animal Care and Welfare Officer, but has failed to ensure that the full range of responsibilities as described in the Guidance on the Operation of the 1986 Act are

satisfactorily performed. This is in breach of Condition 9 (a) of the Certificate of Designation

a. Input at the in-house Ethical Review Committee is offered by the NVS to consideration of Project Licences, although prior discussion with Applicants does not take place. During interview of the NVS, it was confirmed that no discussion or training is given to new staff by the NVS on the recognition or implementation of humane end points and no input given to staff/Licensees training in Schedule 1 killing of animals. This is out of compliance with Section 4.63 of the Home Office Guidance on the Operation of the 1986 Act
    i. "At a scientific procedures establishment, the Named Veterinary Surgeon should also: advise licensees, applicants and others on how to implement the principles of replacement, reduction and refinement. In particular, to advise about the impact of experimental procedures on the welfare of protected animals; the recognition of pain, suffering, distress or lasting harm; general and experimental surgical techniques, and post-operative care; appropriate methods of general anaesthesia, analgesia and euthanasia; strategies to minimise the severity of protocols, including the recognition or implementation of suitable humane end-points;
    ii. be familiar with the main provisions of the project licences in use, in particular the adverse effects expected for each protocol; the means by which they are to be avoided, recognised and alleviated; and the humane endpoints to be applied;"

b. During interview with the NVS, he stated that on no occasion has the NVS required a mouse on potency study to be humanely killed; and he does not review the training of staff or their competency in undertaking procedures.

c. Following discussions with the RCVS, a HO formal request to the NVS for evidence of Continued Professional Development over the preceding five years failed to provide appropriate and satisfactory evidence of such commitment. The Guidance on the Operation of the Act (section 4.61) requires a Named Veterinary Surgeon to
    i. "keep abreast of developments in the use of laboratory animals, including the selection of appropriate animal models;
    ii. be able to advise on methods of reducing adverse effects on animals by refinement of experimental techniques and husbandry methods; and
    iii. be able to contribute to ensuring that consideration is given to the use of non-sentient alternatives."
d. On at least one occasion both the NVS and the veterinary surgeon appointed by the NVS to serve as his deputy were alleged by the BUAV investigator to be not contactable when veterinary advice/intervention was requested over a weekend period. This allegation has been refuted by the NACWO at the Establishment who

claims that the deputy veterinary surgeon was available for the period in question. No conclusive evidence has been received to confirm or refute this allegation.

e. Overall, by failing to appropriately monitor and advise on the application of humane end points in mice bioassays the NVS has not discharged his duties to a reasonably satisfactory level. Additional adverse animal welfare burdens occurred due to this failing.

f. The responsibilities of the NACWO include day to day supervision of work in the animal facilities, maintenance of appropriate records as required by the SoS and ensuring that daily checks are made on all animals held in the establishment.

g. Concerns over the supervision of Schedule 1 killing are addressed elsewhere in this Review, although such concerns do not extend to the adequacy of record keeping by the NACWO for such work. The records were inspected and were complete an dup to date. Daily animal checks were made and recorded in an appropriate format.

h. The NACWO had been on extended leave recently and had delegated some responsibilities to Senior Animal technicians – notably training and assessment of new staff.

***Recommendation 2***

That the Holder of the Certificate of Designation at Wickham Laboratories be required to demonstrate to the Secretary of State that the full range of responsibilities expected of the Named Persons nominated under his Certificate of Designation are being discharged. In particular the Certificate Holder must assure the Secretary of State that the duties required of his Named Veterinary Surgeon (as laid out in the Guidance on the Operation of the Act and by Annex K – NVS Guidance - of The Royal College of Veterinary Surgeons Guide to Professional Conduct) are being met. Formal infringement action should be considered with respect to lack of compliance with Conditions 9A and 17 of the Certificate of Designation.

***Recommendation 3***

That the Holder of the Certificate of Designation at Wickham Laboratories be required to ensure, and to provide evidence of, regular and effective liaison between those with responsibilities under the Act and with others who have responsibility for the welfare of the protected animals kept there.

## *d. Did Designated Persons, licensees and other staff at the DUE fully and appropriately discharge their responsibilities under the Act?*

74. The majority of staff interviewed at Wickham Laboratories during this review, including the Certificate Holder, Project Licence Holder, Personal Licensees and NACWO have been fully cooperative and responded fully to requests for further data.

75. The Holder of the Certificate of Designation, as stated above, is considered to have incompletely discharged his duties under the Act in that his appointed NVS did not appropriately provide leadership, guidance and advice to others at the Establishment

    *a.* There is concern that the training of staff may be too rushed or may focus inadequately on some important aspects of animal welfare during studies. Statements made by Wickham staff (at interview on 2nd November 2009 and subsequently) repeatedly indicate that the prevailing advice from the PPL holder and senior technicians was to positively encourage staff to err on the side of 'not choosing euthanasia' for mice on study so that 'results are not biased' by such mortality data. If the technicians were in doubt as to whether the animals would survive to the next observation point the advice was to leave the animals until the next observation.

76. The advice given, and practices followed with respect to implementation of earlier humane end points in the mouse potency bioassays is in breach of Condition 6 of the Project Licence. The consequences of such a breach were that animals were subject to unnecessary and additional suffering and left to die *in extremis* rather than being humanely killed at an earlier time.

77. Allegations that staff were practicing injections on live mice may be considered to also be an example of rushed training. No examples of 'practice' injections were identified, although staff holding newly obtained Personal Licences are expected to undertake injections during active studies. Such Licensees are under direct supervision of a more experienced member of staff until they are deemed to be competent in the technique. No evidence was found that group sizes or animal numbers were increased to provide a population of animals for 'practice' injections.

78. No use is made at Wickham Laboratories of injection technique training or assessment using mouse cadavers, which might be considered good practice at Designated Establishments.

79. In their own published data and from examination of study records for the mouse potency bioassay, the proportion of mice humanely killed has been as low as 0% and is typically around 20%

80. A summary of the data for three bioassays directly observed on a visit during this review is given below. A comparison with three assays run in the preceding month, with cumulative observations to the same time point within

the assay, is also included. Although staff claim to have changed nothing in the criteria or in their practices, there was a marked increase in the proportion of mice humanely killed in the three assays still running compared to similar assays from two weeks previously (Prior to initiating this review)

| | FD | | HE | |
|---|---|---|---|---|
| **Study Number** | **Number** | **% of animals that did not survive** | **Number** | **% of animals that did not survive** |
| 117/1 | 31 | 54.4% | 26 | 45.6% |
| 117/2 | 29 | 50.9% | 28 | 49.1% |
| 117/3 | 25 | 65.8% | 13 | 34.2% |
| | | | | |
| **Total** | 85 | 55.9% | 67 | 44.1% |
| | | | | |
| **Comparator data** | | | | |
| 95/5 | 41 | 74.5% | 14 | 25.5% |
| 95/1 | 33 | 73.3% | 12 | 26.7% |
| 95/2 | 32 | 69.6% | 14 | 30.4% |
| | | | | |
| **Total** | 106 | 72.6% | 40 | 27.4% |

(FD = Found dead; HE = Humanely euthanased)

81. Records summarised from mouse potency studies conducted in 2003 on another botulinum containing product at Wickham have shown similarly high ratios of animals 'found dead' as compared to 'humanely euthanased'. This work was not conducted under the authority of the current Project Licence and used a reduced frequency of clinical observations at approximately 4 hourly intervals.

| **Study Ref (2003)** | **No** | **Survived** | **% of total animals** | **FD** | **% of total animals** | **HE** | **% of total animals** | **FD as % of animals that did not survive** |
|---|---|---|---|---|---|---|---|---|
| | | | | | | | | |
| 186701 | 200 | 78 | 39.0% | 121 | 60.5% | 1 | 0.5% | 99.2% |
| 273636 | 200 | 56 | 28.0% | 143 | 71.5% | 1 | 0.5% | 99.3% |
| 292998 | 200 | 88 | 44.0% | 112 | 56.0% | 0 | 0.0% | 100.0% |
| 472117 | 200 | 67 | 33.5% | 128 | 64.0% | 5 | 2.5% | 96.2% |
| | | | | | | | | |
| Total | 800 | 289 | 36.1% | 504 | 63.0% | 7 | 0.9% | 98.6% |
| | | | | | | | | |

(FD = Found dead; HE = Humanely euthanased)

82. The Standard Operating Procedure (SOP) produced by senior staff at Wickham Laboratories states: "*Any mice showing very severe symptoms, such that the experienced qualified technician considers that the animal will not survive until the next observation period, must be killed by a Schedule 1 method. Remove any dead animals from the mouse box at each observation period. Record any mice removed from the assay on the Mouse Observation and Population Record (FT082): enter the number found dead and/or culled, date, time, and initials*" This instruction is not in line with the verbal advice given to technicians by the PPL holder and other staff. The advice, repeated during interview at the Establishment, from the PPL holder is to leave mice until the next observation period if staff are uncertain over its viability. The BUAV investigator stated that on occasions (during night time observation periods) she fulfilled the role of being the more senior of the two technicians on duty at that time.

83. The Project Licence states in Section 18 *"Mice are observed at regular intervals, the frequency increasing to coincide with increased effect of the toxin on the mice. Mice showing severe symptoms of paralysis, including difficulty with breathing, wasp waisting and cyanosis are considered unlikely to survive until the next observation period. Animals, where these symptoms are clearly observed and it is considered the animal will not survive to the next observation period will be killed."*

84. Additional specific steps are laid out in the Section 19 protocol for this test: *"At each observation any mice showing very severe symptoms will be killed by a Schedule 1 method. Any dead animals will be immediately removed"* and "*All animals except possibly those in the very low dose groups will show typical signs of Botulinum Toxin to some degree; this includes difficulty with breathing (wasp waisting) and some limb paralysis. The effects are controlled by observing the animals frequently and killing by a Schedule 1 method any animals that an experienced qualified technician considers will not survive until the next observation period. Regular observation is commenced as soon as the mice in the highest dose group start to show severe symptoms and then continued on a regular basis with the highest frequency when symptoms are at the most severe. For example currently approximately hourly observations are made during the high risk period and the situation regularly reviewed."*

85. In practice, an 'experienced qualified technician' was taken to mean an animal technician who may also hold a HO Personal Licence and has experience of conducting the specific regulated procedures as authorised within a PPL. Under guidance from the PPL holder, there was a liberal application of the criteria detailed above such that mice were not killed despite being unlikely to survive to the next observation period.

86. The time intervals noted on laboratory records for observations of animals in mouse bioassay studies varied from slightly under 1 hour to over 2 hours – with an interval agreed in the project licence of approximately 1 hour during the high risk period. Some variability occurs due to the number of mice being

observed by individual Licensees, although this would be minimised by allocating additional resources to the times of more intensive observations.

87. Whilst 'approximately hourly' permits a reasonable tolerance in the expected intervals between clinical observations and the taking of any necessary actions, the irregular and at times lengthy intervals which were practised during the mouse bioassays exceed this tolerance. In the context of these studies, an extended observation interval, outside that agreed in the project licence for the high risk period and potentially causing unnecessary suffering, might be considered to be lack of compliance with licence authorities

88. As noted in some HOI Visit Reports, conducting even more frequent observations may not accurately detect mice which were approaching death – the Inspectors following a PIL holder through their observations and noting additional animals requiring euthanasia only minutes after the same animals had been considered capable of surviving to the next (1 hour) observation period.

89. Given the rapid deterioration in animal health which may occur in this assay, it is essential that the guidance being followed with respect to earlier intervention (and euthanasia) is clear and is consistently applied.

90. An effort to improve the application of humane killing during the conduct of the mouse potency bioassay was initiated during 2008 at the suggestion of the HOI and has been conducted at Wickham Laboratories. The outcomes were discussed during 2008 with the HO Inspector and at the Establishment's Ethical Review Committee. Following changes to the criteria for applying the humane end points, it was shown that identifying mice which were considered 'doubtful' (i.e. those for which the PPL holder's previous advice had been to leave until the next observation point) did not result in culling animals which would have survived to the end of the test.

91. The Establishment and the PPL holder have failed to apply this refinement to their subsequent conduct of these assays.

92. More recent data for the three months November 2009 – January 2010 is summarised below for the 15,344 mice placed on test in the mouse potency bioassays.

| Study Ref | No | Survived | % of total animals | FD | % of total animals | HE | % of total animals | FD as % of animals that did not survive |
|---|---|---|---|---|---|---|---|---|
| 65 studies | 15,344 | 7,267 | 47.4% | 5,480 | 35.7% | 2,597 | 16.9% | 67.8% |

93. The above data suggest that refinement is possible leading to earlier application of humane killing and fewer animals being 'found dead.

94. The percentage of mice found dead (as a percent of all deaths on study) during potency studies has ranged from around 50% to 100%. Sustained attention to frequent and critical clinical observations should improve both the wide range of 'percentage found dead' animals and should allow for sequential improvement in this ratio by better guiding the timing and content of clinical observations.

95. Progress made on refinements to humane end points on the mouse bioassay was not followed up by the Project Licence Holder. Work which had been completed during the pilot study was discussed at the Establishment's Ethical Review meeting, but had not focussed on the changes which should be made and need for revision of day to day practices.

96. Personal Licensees were observed undertaking their work, and have been further reviewed in those video clips selected and provided by BUAV. The work included regulated procedures and killing by Schedule 1 methods.

***Recommendation 4***

That the Holder of Project Licence PPL 70/6936 *"Regulatory Testing of Biological Toxins"* write to explain how he will improve his supervision of those conducting work on this Project Licence to ensure that early and humane end points are consistently applied at all times. Formal infringement action should be considered with respect to lack of compliance with Condition 6 of his Licence

## *e. Was oversight and action by the Home Office in general, and in particular by ASPI, appropriate to the DUE and its work?*

97. Applications for Project and Personal Licence authorities were assessed and processed appropriately in line with current documented systems of work.

98. During the period 2005 to 2009, Home Office Inspectors appointed under ASPA visited the Wickham Establishment on 25 occasions. These visits have been at the rate of approximately 4–5 inspections *per annum*, with a small number (4 in total out of 25) being visits made jointly with another HOI.

99. The oversight by the Home Office of Wickham Laboratories extended to 25 visits of inspection over a five year period, coupled with assessment of applications for Personal and Project Licences during that time. This level of oversight is considered appropriate for an Establishment of this size, although not all issues raised by Inspectors were followed through to completion by the duty holders at Wickham.

100. The DUE was assessed on the basis of number of Project and Personal Licences, species used, severity bands of the Project Licences and recent compliance history to represent a moderate/upper moderate risk. Factors which would have raised the risk profile would have been use of any of the species afforded special protection under ASPA, (dogs, cats, equines and non-human primates), involvement of surgical or other additional regulated procedures and current/recent history of infringements.

101. There have been 4 changes of HO Inspectors assigned responsibilities for Wickham Laboratories at different times during this period (2005 -09). This frequency of change is higher than typical within the Inspectorate, where an individual Inspector will typically take lead responsibility for a DUE for period for 4 – 5 years. Each Inspector has reported inspections during the period, with visit durations from 0.75 – 2.50 hours. (A single report documents a 3.00 hours visit, but a significant part of this was taken up with discussions on scientific presentations to an external group of scientists)

102. The Inspectorate has an internal guidance document on the handover of designated places which indicates that from the Home Office perspective the intention must be to ensure that any handover takes place as efficiently and effectively as possible and that the formal documentation will clearly define:

a. the status of the place,
b. current issues and problems
c. initiatives in order that the incoming inspector can build on the activity and achievements of his or her predecessor.

103. It is apparent that such formal documentation was not always in place for this Establishment; the exceptions occurring when one HOI was re-assigned the responsibility following resignation of an Inspector (and therefore arrived with prior experience of the Laboratories), and on one occasion when the ‘out-going’ HOI was absent on sick leave.

104. The higher than usual rate of change of Inspectors, caused in part by extended sick leave and by resignations, contributed to incomplete follow through on some of the initiatives at the Establishment.

105. The average duration of an inspection visit was just under 2 hours (1hour 50 minutes). The facilities comprise 12 animal holding rooms and three procedure rooms (including the pyrogen suite as a procedure room). Not all animal rooms are occupied on all occasions. This duration of visit is not considered unusual for inspection of establishments of this size, assuming that no significant findings are made during the visit.

***History of Home Office Inspection Visits and Findings***

106. The Establishment has been regularly inspected by ASPI throughout its time of designation under ASPA. Changes in ASPI staff have meant that six different Inspectors have been assigned oversight of Wickham Laboratories in the period 1993–2009.

**107.** Working within a risk balanced programme of visits it is not possible for Inspectors to observe all procedures and systems of work (such as killing by Schedule I methods) at each Establishment. The preponderance of inspections being unannounced (i.e. with no prior notification to the Establishment) means that not all aspects of work on each Licence may be seen at a single Inspection. Over the course of time, all significant stages of work conducted at Wickham Laboratories were inspected during ASPI Inspections.

**108.** The aspects of the work inspected include mouse bioassay injections and clinical observations; rabbit pyrogen testing; killing by a Schedule 1 method (cervical dislocation); animal husbandry; design of studies; condition of facilities; and environmental controls

109. Findings of Inspectors with respect to the monitoring of humane end points in the botulinum studies have indicated variability in the application of an appropriate end point:

a. "*one mouse was determined to warrant euthanasia which was immediately carried out satisfactorily by Mr W (a PILH). As this (was) seen between observation points and because Mr W acted independently and quickly to resolve the issue, I do not consider there to a problem with compliance or monitoring. Otherwise all animals in satisfactory condition and the regulated procedures observed above were carried out competently with minimum suffering to the animals*" (Nov 2005)

b. "*I was satisfied that the animals undergoing potency testing were being monitored appropriately and the adverse effects adequately managed*." (January 2006)

c. "*Perusal of the monitoring schedule indicated that observations were not always conducted at the hourly intervals stated, and few animals were humanely killed rather than being found dead*." (October 2006)

d. One report includes a statement that appears to confirm that the prevailing system may fail to detect animals in need of humane killing: "*I found a number of mice dead or approaching death (loss of righting reflex) whilst on the biological toxin potency assay. I raised this with Mr W and he offered to decrease the interval between observations. Records show that monitoring is adequate and that the mice found were soon to be observed hence the proposed increase*" (July 2006)

110. Through 2006, 07 and 08, the Inspector continued to challenge the use of non-randomised cage/dose groupings and to suggest methods for improving the ratio of found dead: humanely killed. By April 2008, little progress had been made on either of these: "*Mr Z (a PILH) reported that no progress had been made since my last visit in blinding studies or refining the humane end point, because of lack of time. This was disappointing. Mr Z is keen to progress these and the move towards the lower severity flaccid paralysis assay but has simply been too busy to do anything about it*." (April 2008)

111. A proposal for refining the mouse bioassay was agreed with the PPL holder: "*Visit to follow up on initiatives to refine the Dysport assay. Mr Y (PPLH) reported that the initial study had identified that the current scoring system did not result in culling animals which would have survived, but did fail to pick up all those which would die and could be made more sensitive. He hopes to introduce a simple 'on/off' system whereby animals exhibiting one or more specific signs are culled, making the scoring less subjective and culling animals at an earlier end point. Progress has also been made in blinding the scoring, and in automated reading of the lower severity flaccid paralysis assay: this needs to be followed up*." (August 2008)

112. Current data do not demonstrate ongoing practices have continued to change for the better. 80% of study related deaths are still "found dead" and local DUE guidance is to err on the side of 'not humanely killing' if in doubt.

113. During 2005 advice was given by HOI and acted upon by Wickham to provide larger caging for stock rabbits; intra-peritoneal injection of mice and pyrogen testing of rabbits were directly observed as were animals at varying stages of the botulinum potency bioassay. On one occasion the HOI required one mouse to be humanely killed while on bioassay due to the severity of suffering. This occurred between the approximately hourly observation times and was not deemed to be evidence of non-compliance.

114. An issue of poor compliance with Code of Practice requirements for environmental control was also discovered in 2005 by the HOI and recommended for prompt resolution by the PCD Holder.

115. During July 2006 killing of mice by Schedule 1 method (cervical dislocation) was directly observed and reported as competently undertaken.

116. Advice was also given to animal care staff to ensure careful and complete replacement of cage tops in order to prevent tail damage. The issue of adding a group of animals to the mouse biological toxin potency assay in order to 'validate' a new technician was discussed. The HOI view was that it was not considered to be a permissible purpose under ASPA and that due consideration be given to non-animal alternatives and/or training using cadavers etc prior to I technical staff conducting such assays.

117. During mouse bioassay, the cages were placed on the racks in their treatment groups and the trials were not blinded, and the HOI raised concerns that there was considerable potential for observer bias in the monitoring. The monitoring schedule also indicated that observations were not always conducted at the hourly intervals stated, and few animals were humanely killed rather than being found dead. It was noted that the previous HOI had discussed this with the group at length but it was not clear that they have fully understood, or that a suitable distress scoring system had been identified to implement humane end points.

118. During 2007 one mouse on bioassay was found dead on inspection, another was culled by the technician during the HOI visit. The possible ways to improve the predictability of the observations to allow for culling rather than leaving animals to be found dead was discussed with the NACWO. Although the technicians are experienced, the observation system used did not appear particularly systematic, although several objective criteria were used.

119. On a further visit in 2007, the HOI noted that several mouse bioassays were in progress. *"These had just been scored, however I found one animal moribund in a high dose group, and the NACWO arranged for it to be killed, and for the scoring to be re-done in that room. At least four more animals were humanely killed following this second round of scoring. This serves to emphasise the difficulties associated with this test: the output is survival, yet clearly implementing humane end points is to be preferred to death. The subjectivity of the distress scoring system remains an area in need of improvement"*

120. Improvements in provision of floor pens/exercise cages for rabbits were noted and rabbit pyrogen testing continued to be noted by the HOI to be satisfactory. Also discussed with the Project Licence Holder was the scoring method used for determining the mouse bioassay end point. Cages are all marked in a way that identifies the dose received. Therefore, the scoring system, which is rather subjective, is likely to be biased, possibly overestimating the severity in the high dose groups and underestimating it in the low dose groups. It was suggested to the PPL holder that the cages be

marked such that those doing the scoring did not know which group was which, to reduce bias.

121. Later in 2007 during a joint Inspection, two HOIs visited while two mouse studies were underway. The records indicated that more animals were found dead than humanely killed, and that periods between scoring were rather variable. Scoring had just taken place, yet the HOIs identified a further 3 or 4 dead animals and others that were near the end point. This was discussed at length with the Personal Licensee involved. The criteria used for determining the end point continue to be subjective, and since the scientific end point is survival to 72 or 96 hours there is reluctance to kill animals too early to avoid skewing the data. The HOIs also discussed again the potential for *in vitro* tests with the PPL holder – progress was reported as being made towards validating an *in vitro* test although no evidence was provided of such progress.

122. During 2008 detailed discussions were held with the PCD holder and PPL holder over positive steps to improve the outcome of the mouse bioassay with respect to animal welfare. A pilot study was proposed by the HOI to determine whether changes in criteria for Schedule 1 killing within study would skew the data/results. This was taken forward to the Establishment's Ethical Review Process.

123. During a further unannounced visit, a PIL holder reported that no progress had been made since the last HOI visit in blinding studies or refining the humane end point, because of lack of time. At a subsequent visit to follow up on initiatives to refine the mouse bioassay the PPL holder reported that the pilot study noted above had identified that the current scoring system did not result in culling animals which would have survived, but did fail to pick up all those which would die. The PPL holder was reported to be willing to take this forward.

124. Improvements in the conduct of the rabbit pyrogen testing were proposed by the HOI, and implemented by the Establishment. These included the use of topical cream and finer gauge needles to minimise discomfort from injection into rabbit ear veins.

125. The Inspectors have repeatedly given advice to Licence holders at Wickham Laboratories and have identified areas of concern in the conduct of work under Licences held there. Examination of the Visit Reports indicates however that follow up to previously raised concerns has been inconsistent on some occasions.

126. When an Establishment is 'handed over' to another Inspector, Guidance from ASPI suggests that previous Visit Reports, along with recommendations or observations contained therein, should be reviewed by the outgoing and incoming Inspectors and follow up actions should be taken. This has not always been demonstrably undertaken at Wickham Laboratories.

127. The ‘handover’ Guidance states that whilst a joint visit between the outgoing and incoming Inspectors may be helpful, it is not always essential. In the case of Wickham Laboratories, one Inspector re-assumed responsibility for the establishment and no specific joint visit was made, and on the most recent changeover the outgoing Inspector entered a period of prolonged sick leave immediately after a joint visit, prior to agreeing any necessary follow ups with the incoming Inspector.

128. With respect to this particular Establishment, several areas may be suggested for further consideration by the Home Office Inspectorate:

a. More targeted review of Visit Reports from this DUE so that where recommended, follow up actions can be appropriately taken;
b. More frequent review of the risk profile offered by the DUE and adjustment of the Inspection pattern/content to more adequately reflect this and to permit Inspection of critical phases of studies;
c. When DUEs are assigned to another HOI, the provision of previous Visit Reports and discussion of outstanding actions for follow up are a high priority. It was noted that joint handover visits were not always completed at each period of change at Wickham Laboratories.

***Recommendation 5***

That ASPI management should review the oversight of this Designated Establishment against the existing guidance to Inspectors, including guidance on handover of Establishments to ensure that issues are now being appropriately followed through.

## *f. Was the work done at Wickham - including Schedule 1 killing of animals - completed to good contemporary standards?*

129. Killing by Schedule 1 methods was observed on several occasions, both directly by Inspectors at the Establishment and through the submitted video clips. This included killing of individual animals and video records of group ($CO_2$ exposure) euthanasia.

130. The killing of mice by cervical dislocation is a method listed in Schedule 1, but concerns exist over the appropriateness of conducting this on corridor floors as opposed to at normal working height and of the competency with which it was carried out:

a. There are occasions when it is alleged that the attempted cervical dislocation produced other fracture/dislocations; broken backs. This was not observed to be the case on any inspection visits made by HOIs. Some staff admit that this may occur and that the amount of training to develop good technique is limited. Since cervical dislocation produces rapid ascending and descending trauma the over-riding goal is to achieve efficient cervical dislocation.
b. Limited research data exists on the incidence of other vertebral dislocations occurring during attempted cervical dislocation, but it is reported that thoracic or other sites may also be affected in a small number of cases.
c. Video evidence shows mice with movements after Schedule 1 killing by one staff member and whilst it is not possible to state with certainty that this was due to poor technique, the extent and duration of movement is more than expected.
d. In addition to failures of technique, the routine procedure which forms the basis for training in cervical dislocation of mice has been to remove the mouse from its home cage, take it outside the animal room to the corridor and to complete the killing on the floor of the corridor. No reported use of, or observation of, this procedure was made by any of the Inspectors in their Visit Reports.
e. Killing on the corridor floor is an unacceptably poor system of work. The unbalanced angle of work will produce difficulties in achieving the correct alignment of pressures/traction for a prompt, humane and complete kill.

131. During interview with several members of staff at Wickham Laboratories, a rationale for undertaking cervical dislocations in the corridor was given:

a. The Code of Practice for Schedule 1 Killing advises that animals are removed from others in their group and that killing should not take place in the presence of the other cage mates;
b. Technicians and management considered that removal from the home room into the corridor would be in line with this aspect of the Code;
c. The NACWO and management at Wickham Laboratories did not challenge the practice.

132. The use of a carbon dioxide (CO2) chamber for euthanasia of groups of mice is an acceptable method under Schedule 1. A SOP exists at Wickham Laboratories for use of a specifically designed single chamber located within the animal facilities.

133. The SOP requires animals, up to 60 in number at any one time, to be removed from their home cages and to be simultaneously exposed to rising concentrations of CO2. Animals will be mixed within the chamber from different home cages and will include healthy and adversely affected animals at the end of studies.

a. The chamber has internal floor dimensions of 44 x 30 cm, with a height of 25 -28 cm (the difference being produced by a slight curving of a false floor). Effective floor area is therefore approximately 1,320 sq cm

b. Groups of up to 60 live mice are placed into the chamber and CO2 introduced at a lower then full rate to induce unconsciousness followed by death. Animals are removed after a minimum exposure period of eight minutes and death is confirmed by checking for rigor mortis.

134. Justification, other than historic practice, for permitting the use of a group size of up to 60 mice at a time could not be provided by the staff or management. One opinion offered was that "*60 would make a monolayer in the chamber*", i.e. would completely fill the floor. This is not good practice in that all mice should be able to move and change posture within a CO2 chamber

135. On at least two occasions it was alleged by BUAV that more than 60 (up to 65) mice were placed simultaneously within the CO2 chamber.

136. One video clip shows a technician (the BUAV investigator) carrying a box of mice and noting that after being removed from the CO2 chamber it was unconscious but still breathing. The animal was immediately killed by another technician by cervical dislocation. This supports in part a further allegation that confirmation of death (a requirement of Schedule 1 procedures) was only performed in a superficial manner; the main activity of the technician charged with such work being to confirm the numbers of animals live/dead for the study records.

137. Mice which died had reached death in one of three ways:

a. Those found dead during conduct of studies. Clinical examination of the animal to confirm cessation of breathing and any other sign of life was followed by removal from the home cage and placing in a separate cage maintained specifically for that purpose. A second Licensee/ Technician then examined and counted the decedents. No additional procedure or check was applied to these animals.

b. Mice killed individually by cervical dislocation during the course of studies. These mice were removed from their home cages and killed (as described elsewhere in the Report) by cervical dislocation, a method specified in Schedule 1 to the 1986 Act. Visual checking and counting of animals which had been killed in this manner was conducted by a second Licensee or Technician. No additional procedure or check was applied to these animals.

c. Mice killed in a group by exposure to rising concentrations of carbon dioxide. As described elsewhere in the report, mice surviving to the end of studies were killed in groups within a carbon dioxide chamber. On completion of the procedure (documented and updated by Wickham Laboratories) mice were removed from the chamber and examined by a second Licensee or technician to confirm death. Paragraph 136 of this report documents an instance when one mouse was detected to be still breathing after removal from the carbon dioxide chamber and was immediately killed by cervical dislocation

138. **Conclusion:** The conduct of Schedule 1 killing of mice at Wickham Laboratories is inconsistent, at times incompetent and requires improvement.

139. The housing of mice at Wickham is routinely in groups within purpose made cages held on mobile racks. Feed and water are provided through hoppers/bottles held in the cage lid. No inspection report identified deficiencies in housing or husbandry of the animals.

140. Provision of an in-cage source of water to severely impaired mice, such as those on botulinum bioassay potency studies, could be improved since the developing paralysis may impede movement. Water pouches and hydrogel (transport) devices would provide such an alternative.

***Recommendation 6***

A comprehensive review of Schedule 1 killing to be undertaken by the Holder of the Certificate of Designation in order to achieve consistent improvement in both the processes and the outcomes. This review to include:

a. All staff currently registered as trained and competent in Schedule 1 killing to be assessed and where necessary re-trained, using accredited trainers;

b. Stopping the use of corridor floors for the conduct of Schedule 1 killings and provision of appropriate work-tops at normal working height for individual animal kills;

c. Revision of the SOP for use of the CO2 chamber so that all mice should be able to move and change posture within a CO2 chamber when the chamber is filled;

d. Ensuring the establishment is adequately equipped to euthanase animals efficiently and humanely using carbon dioxide;

e. Revision to the SOPs and improvement of supervision such that all steps during Schedule 1 killing – including positive confirmation of death as required by the Act – are fully and consistently completed

The conduct and outcomes from this review must be to the satisfaction of the Secretary of State and completed without delay.

## 5. DUE status

### *a. Certificate of Designation, Project Licences and Personal Licences held*

141. Wickham Laboratories is a Designated User Establishment (DUE) under The Animals (Scientific Procedures) Act, 1986. The holder of the Certificate of Designation (PCD) is the Technical Director who reports to the Managing Director

142. The Named Veterinary Surgeon (NVS), who is also a major shareholder in the Company, qualified as a veterinary surgeon in 1950 and has held the position of NVS since 1986, although he has not undertaken the now mandatory training for newly appointed NVS. (His appointment was made, and has been held continuously since before the introduction of NVS mandatory training.)

143. The NVS is also the Managing Director of the Laboratories, to whom the PCD holder reports

144. The NACWO is an experienced animal technician and has worked at Wickham Laboratories for over 17 years

145. The DUE has 3 Project Licences (PPLs) in force (held by two Project Licence Holders) and 11 Personal Licences (PILs).

### *b. Procedures undertaken and function of the Ethical Review Process*

146. Around 89,000 regulated procedures are reported annually by the Establishment within their Returns of Procedures. The majority of these use mice, mostly in botulinum potency assays.

147. Of the approximately 1,800 regulated procedures reported annually by the Establishment which use rabbits (pyrogen testing), a majority are classified as authorised re-use.

148. The Ethical Review Process (ERP) is chaired by the Certificate Holder (who is also a PPL holder) and meets at approximately quarterly intervals. The NACWO acts as Secretary to the ERP and an in-house member of staff holds the role of Lay Member. The NVS and other PPL holder also attend the ERP

149. The Lay Member is invited to informally visit the animal facilities as they wish, although this not a frequent occurrence.

150. Minutes and Agenda for each ERP meeting are brief and address issues with Licences, feedback from any visits by the HO Inspector and reports of conferences/meetings attended by staff.

## *c. Potential for Conflict of Interest*

151. Concern exists that an actual or perceived conflict of interest may exist between the various positions held by the NVS at Wickham

a. As NVS, provider/ensurer of provision of veterinary care to animals
b. As NVS, provider of advice to scientists, Licensees and others holding positions and responsibilities under the 1986 Act
c. Major shareholder and co-owner (with his wife) of Wickham Laboratories
d. Managing Director of Wickham Laboratories
e. Owner of animals used in programmes of work authorised under the 1986 Act

152. Additionally, certifying one's own animals is against recital 3 of the 12 Principles of Certification (Annex D of The RCVS Guide to Professional Conduct). This has been discussed with the RCVS this may be open to other interpretations in a case such as this. (as opposed to certifying one's own companion animals)

153. A further potential for conflict of interest occurs between the different roles of the Certificate Holder and his nominee for the position of NVS. As Holder of the Certificate of Designation for the DUE and as a Project Licence Holder he must expect to receive comprehensive advice and guidance on his obligations under ASPA. By placing in the NVS post his own employer there may be occasions when decisions are influenced by those employment roles and not by the expected responsibility of a Certificate Holder.

154. Two sets of Guidance exist to inform individuals who may hold authorities under the 1986 Act. These are the Guidance on the Operation of the Animals (Scientific Procedures) Act, 1986 –published by the Home Office, and Guidance for Named Veterinary Surgeons published as Appendix K to the RCVS Guide to Professional Conduct by the Royal College of Veterinary Surgeons.

***Home Office Guidance***

155. Section 3.17 of the Guidance states that the Act makes provision for a number of individuals to assume responsibility for different aspects of the well-being of protected animals at designated establishments: certificate holders, project licence holders, personal licence holders, Named Animal Care & Welfare Officers, and Named Veterinary Surgeons.

156. In Section 3.18 it confirms that the contractual relationships between, and other responsibilities of, these individuals can create conflicts of interest. There may also be occasions when one individual legitimately fulfils more than one of these of roles. For example, in most cases, project licence holders will hold, or will have held, a personal licence. For these reasons, the Secretary of State normally requires that, for any group of protected animals, at least three individuals should fill these five roles.

157. Section 3.19 indicates that when a Named Veterinary Surgeon or Named Animal Care & Welfare Officer has (under any other of the statutory roles) a substantial interest in the scientific outcome of a programme of work, alternative provision should be made for the veterinary or welfare oversight of the animals in question.

***RCVS Guidance (Appendix K to the Guide to Professional Conduct)***

158. Paragraph 16 of the RCVS Guidance states that, where the NVS also holds a project licence, another veterinary surgeon must be agreed with the Home Office as responsible for providing independent veterinary advice regarding the health and welfare of the animals involved. If there is any other significant conflict of interest, the NVS should consider the need for independent veterinary advice.

***Considerations arising from Guidance Notes***

159. The current review of allegations made against Wickham Laboratories appear to encompass a situation envisaged under the HO Guidance
*"3.18 The contractual relationships between, and other responsibilities of, these individuals can create conflicts of interest."*
and the RCVS Guidance,
*"If there is any other significant conflict of interest, the NVS should consider the need for independent veterinary advice"*

160. Considerable concern was expressed at the meeting of the Animal Procedures Committee on 3 February 1994 that, for example, the managing director of an organisation might be responsible for discharging either of the named veterinary surgeon or day-to-day care person roles. It was noted that there are difficulties in formulating specific rules concerning potential conflict when a person with a major financial interest in any operation of an establishment where animal work is carried out may have one of these two

roles which have a particular importance in protecting animal welfare under the Act.

161. The then Chief Inspector concluded that "*whilst it is acknowledged that there is no mechanism whereby inspectors will necessarily be made aware of the financial interests of all persons appointed under A(SP)A, they should seek to divorce the roles of day-to-day care person and named veterinary surgeon from such major interests where they are known*".

162. It has not proved possible to determine how frequently or to what depth enquiries were specifically made by the Inspectors as to the degree of financial interest held by the NVS at Wickham Laboratories. Records examined from the period following the previous investigation (1993) show that changes to management and training at the Establishment were not deemed by ASPD at that time to be unsatisfactory.

163. Advice was given in 1993 that 'E Division' (the fore-runner to the current ASPD) would set up and maintain a register on which would be held information relating to actual or potential conflicts of interest which came to the attention of Inspectors. No evidence was found during this review that such a register was initiated and no such register currently operates within ASPD or ASPI.

164. **Conclusion:** It would appear that the complex contractual relationships which pertain at Wickham Laboratories with respect to the current NVS have created at least a perceived conflict of interest.

***Recommendation 7***

That the Home Office should give further consideration to situations from which conflicts of interest may be perceived to arise and should publish any revision to their guidance in this area.

# 6. DUE Background

## *a. Compliance History*

165. This Establishment has been subject to exposé investigations from the BUAV in the past and was extensively reviewed in 1992/93 by the Home Office for alleged non-compliances with ASPA. Several members of staff, including the holder of the Certificate of Designation were warned as to future conduct and one employee was removed as NACWO and had his Personal Licence revoked

166. Formal training schemes, improved record keeping and revision to the Standard Operating Procedures in force at Wickham Laboratories were all

required by the Home Office and implemented by the Establishment as a consequence to that review.

167. In a report dated December 1993 the Chief Inspector concluded that the actions required of the Establishment had been completed satisfactorily and that the Holder of the Certificate of Designation should remain in that position.

168. The Laboratories were targeted by members of the Animal Liberation Front in 2003 and resulted in the jailing of one ALF defendant and imposition of a community order on a second ALF person

169. In a series of letters to the Home Office (2005/06) the campaigning group Animal Aid asked why the Home Office continue to license the use of the mouse LD50 test for the testing of such products; why the SNAP-25 endopeptidase assay, used by the National Institute for Biological Standards and Control (NIBSC) to confirm the quality and potency of the finished product, was not yet considered suitable for use by the manufacturers for the tests they conduct; and an open letter requesting that the validation of the SNAP-25 test be ranked as a top priority

170. The replies given by the then Parliamentary Under-Secretary of State, Mr Andy Burnham,MP, stated that:

a. "the mouse LD50 test is currently in our view the method most likely to produce scientifically satisfactory results to meet the requirement that these products are tested to ensure their quality, potency and safety during production, and before they are released for clinical use;

b. that the SNAP-25 test has been internally validated by NIBSC for the tests it carries out at the end of the production cycle, but only when performed at NIBSC under carefully controlled conditions and has not yet been validated for use at the other stages of the testing process or in other laboratories;

c. the validation of a SNAP-25-based test is something we strongly support and that we are committed to moving to less severe testing procedures as soon as it becomes practicable to do so. "

# 7. Conclusions and Recommendations

171. Licence and Certificate authorities to permit work under ASPA 1986 were, and are, in place at Wickham Laboratories

172. The authorities granted by the Home Office under ASPA were issued legitimately and with appropriate reference to current requirements for authorised medicinal products.

173. Whilst alternative assay methods could be developed and validated for the specific products being tested at Wickham Laboratories, this has not yet been completed. The criteria applied through the Marketing Authorisations for medicinal products allow for a manufacturer to state the pharmacopoeial method they choose to employ.

174. The conduct of rabbit pyrogen testing at Wickham Laboratories is in support of the requirements of regulatory authorities for specific medicinal products. The Establishment is well aware of, and participates in, the use of non-animal alternative testing for the detection of pyrogens. At present not all substances can be validly tested for pyrogens using *in vitro* methods

175. The abnormal toxicity testing undertaken at Wickham Laboratories is similarly a requirement of regulatory authorities for either medicinal products, vaccines or medical devices. There is growing acceptance that such testing may be removed from the regulatory requirements for safety testing, provided that sufficient data on batch to batch consistency is provided to the responsible regulatory authorities.

176. The need for product by product validation of alternative tests (in the case of pyrogen detection) or for batch by batch consistent analytical data (in the case of abnormal toxicity) means that animal based studies continue to be a legitimate regulatory requirement.

177. The leadership for such transition, to *in vitro* alternatives, could however be more clearly communicated, e.g. consideration given to use of a specific time period for a Marketing Authorisation during which the manufacturer would be expected to have either validated an alternative or to have conclusively shown that current *in vitro* options were not scientifically valid.

178. The Holder of Project Licence PPL 70/6936 *"Regulatory Testing of Biological Toxins"* must improve his supervision of those conducting work on this Project Licence to ensure that early and humane end points are consistently applied at all times. In particular, refinements should be made to the criteria applied for deciding when to humanely euthanase mice during potency bioassays for botulinum toxin containing products.

179. The conduct of Schedule 1 killing of mice at Wickham Laboratories is inconsistent, at times incompetent and requires improvement.

180. The housing of mice at Wickham is routinely in groups within purpose made cages held on mobile racks. Feed and water are provided through hoppers/bottles held in the cage lid. No inspection report identified deficiencies in housing or husbandry of the animals.

181. Provision of an in-cage source of water to severely impaired mice, such as those on botulinum bioassay potency studies, could be improved since the developing paralysis may impede movement.

182. There is no doubt that the complex contractual relationships which pertain at Wickham Laboratories with respect to the current NVS have created at least a perceived conflict of interest.

## *Recommendations*

### *Recommendation 1*

That the Home Office and other Government Departments consider, perhaps through the Inter-Departmental Group on the 3R's, more clarity in the differing uses of the word cosmetic, so that its use as defined under EU Directive is clearly separated from its use as a descriptor term for certain types of medicinal products.

### *Recommendation 2*

That the Holder of the Certificate of Designation at Wickham Laboratories be required to demonstrate to the Secretary of State that the full range of responsibilities expected of the Named Persons nominated under his Certificate of Designation are being discharged. In particular the Certificate Holder must assure the Secretary of State that the duties required of his Named Veterinary Surgeon (as laid out in the Guidance on the Operation of the Act and by Annex K – NVS Guidance - of The Royal College of Veterinary Surgeons Guide to Professional Conduct) are being met. Formal infringement action should be considered with respect to lack of compliance with Conditions 9A and 17 of the Certificate of Designation.

### *Recommendation 3*

That the Holder of the Certificate of Designation at Wickham Laboratories be required to ensure, and to provide evidence of, regular and effective liaison between those with responsibilities under the Act and with others who have responsibility for the welfare of the protected animals kept there.

### *Recommendation 4*

That the Holder of Project Licence PPL 70/6936 *"Regulatory Testing of Biological Toxins"* write to explain how he will improve his supervision of those conducting work on this Project Licence to ensure that early and humane end points are consistently applied at all times. Formal infringement action should be considered with respect to lack of compliance with Condition 6 of his Licence.

***Recommendation 5***

That ASPI management should review the oversight of this Designated Establishment against the existing guidance to Inspectors, including guidance on handover of Establishments to ensure that issues are now being appropriately followed through.

***Recommendation 6***

A comprehensive review of Schedule 1 killing to be undertaken by the Holder of the Certificate of Designation in order to achieve consistent improvement in both the processes and the outcomes. This review to include:

a. All staff currently registered as trained and competent in Schedule 1 killing to be assessed and where necessary re-trained, using accredited trainers;

b. Stopping the use of corridor floors for the conduct of Schedule 1 killings and provision of appropriate work-tops at normal working height for individual animal kills;

c. Revision of the SOP for use of the CO2 chamber so that all mice should be able to move and change posture within a CO2 chamber when the chamber is filled;

d. Ensuring the establishment is adequately equipped to euthanase animals efficiently and humanely using carbon dioxide;

e. Revision to the SOPs and improvement of supervision such that all steps during Schedule 1 killing – including positive confirmation of death as required by the Act – are fully and consistently completed

The conduct and outcomes from this review must be to the satisfaction of the Secretary of State and completed without delay

***Recommendation 7***

That the Home Office should give further consideration to situations from which conflicts of interest may be perceived to arise and should publish any revision to their policy and guidance in this area.

## 8. References


Adler S et al (2010) **Current Scientific and Legal Status of Alternative Methods to the $LD_{50}$ Test for Botulinum Neurotoxin (BoNT) Potency Testing (Report and Recommendations of the ZEBET Expert Meeting)** In press

Barr et al (2005) **Botulinum Neurotoxin Detection and Differentiation by Mass Spectrometry.** Emerging Infectious Diseases, 11 , 1578 - 64

Ekong T A N, Feavers I M, & Sesardic D (1997) **Recombinant SNAP125 is an effective substrate for *Clostridium botulinum* type A toxin endopeptidase activity *in vitro*** Microbiology (1997), 143, 3337-3347

European Commission (2006) **Statement On The Validity Of In-Vitro Pyrogen Tests,** ECVAM, Mar 2006

Home Office (2000) **Guidance on the Operation of the Animals (Scientific Procedures) Act 1986**

Independent Healthcare Advisory Services (2008) **Standards For Injectable Cosmetic Treatments**

MHRA (2007) UKPAR : **Botulinum Toxin Type A Powder For Solution For Injection (Clostridium Botulinum Type A Toxin Haemagglutinin Complex) PL 06958/0028**

MHRA (2008) **Supply and administration of Botox, Vistabel, Dysport and other injectable medicines in cosmetic procedures**
http://www.mhra.gov.uk/Howweregulate/Medicines/Availabilityprescribingsellingandsupplyingofmedicines/Frequentlyraisedissues/BotoxVistabelDysportandotherinjectablemedicinesincosmeticprocedures/index.htm

Royal College of Veterinary Surgeons (2003) **RCVS Guidance For Named Veterinary Surgeons Employed In Scientific Procedure Establishments And Breeding And Supplying Establishments Under The Animals (Scientific Procedures) Act 1986** Appendix K

Sesardic D and Das R G (2007) **Alternatives to the LD50 assay for botulinum toxin potency testing: Strategies and progress towards refinement, reduction and replacement** Proc. 6th World Congress on Alternatives & Animal Use in the Life Science, 581-585

Straughan D.(2006) **Progress in applying the Three Rs to the potency testing of Botulinum toxin type A.** Altern Lab Anim. Jun;34(3):305-13.

## 9. Those interviewed during this review

### *a. BUAV*

Key personnel were interviewed by the Reviewing Inspector during this review including their undercover investigator.

### *b. Wickham Laboratories*

Authority holders under ASPA and other staff at Wickham Laboratories were interviewed in person. Local SOPs, study documents and data generated from relevant studies were examined

Inspection visits were undertaken to observe the conduct and implementation of humane end points within mouse potency bioassays. Inspection of rabbits on test and within the stock housing facilities took place, but no abnormal toxicity studies were conducted during the period of review visits. Archived data, staff training records and facilities data were examined.

### *c. Home Office Inspectors*

The current and previous Home Office Inspectors from ASPI were interviewed and discussions held with them. Their Visit Report records (and those of earlier HOIs, since retired) were reviewed and recommendations contained within those reports were discussed with respect to follow up and implementation of those recommendations.

### *d. Royal College of Veterinary Surgeons (RCVS)*

The Guidance for Named Veterinary Surgeons is published by the RCVS as a document jointly prepared by RCVS, HO and the Laboratory Animals Veterinary Association (LAVA).

Discussions were held relevant to possible conflicts of interest, appropriate discharge of duties by the NVS and the expected levels of CPD to be attained by an NVS.

### *e. Medicines Healthcare Products Regulatory Agency(MHRA)*

Review of the authorities held at Wickham Laboratories for the testing of pharmaceutical products (batch/lot testing) raised issues requiring clarification from the MHRA on:

i. the legal and supported classifications of botulinum containing products;
ii. the definitions of cosmetics; on the approval and review process for such products

iii. the role of National Competent Authorities in promoting and requiring adoption of non-animal alternative tests during assessment of applications for Marketing Authorisations.

# 10. The Review Group

## *a. Reviewing Inspector,*

**Andrew Coulson, BVetMed, MSc, MRCVS**

Mr Coulson is a Superintending Inspector within ASPI and a veterinary surgeon with 30 years experience of work within the pharmaceutical, research and regulatory disciplines of biomedical science. An Inspector in ASPI since 2001, he has special responsibility for oversight of regulatory toxicology and related programmes of work within the Inspectorate.

In addition to leading within ASPI for a recent external review by the Hampton Implementation Review Team (established to determine the compliance with the Hampton principles by Government Regulators) he has served as a Review Team member for compliance assessment of other Government Regulators.

## *b. Independent reviewers of the report*

### i. Dr John Doe

After having previously worked as a pharmacologist in pharmaceutical industry in the areas of asthma, skin allergy and chronic obstructive pulmonary disease, Dr John Doe joined ICI's Central Toxicology Laboratory in the late 1970s. He initially worked in inhalation toxicology but then managed studies across the full spectrum of regulatory toxicology including chronic, reproductive and developmental toxicology. He became head of project management and business relationships in became Director of CTL, Syngenta in 2003. In 2006 he became Head of Product Safety for Syngenta, bringing together human and environmental safety. He retired from Syngenta in March 2010 ad is now an independent consultant.

He was Chairman of the Scientific Committee of ECETOC from 2006-2010. Dr Doe has published papers in the fields of immunotoxicity, combustion toxicology, reproductive toxicology and risk assessment. He currently serves as a member of the Animal Procedures Committee

### ii. Dr Robert Hubrecht

Dr Hubrecht is Deputy Scientific Director of the Universities Federation for Animal Welfare. He is an ethologist with interests in the animal welfare aspects of the housing and husbandry of laboratory animals, and the ethical issues involved in their use. He has carried out research on the natural history of New World Primates, and on improving the welfare of kennelled dogs.

He has chaired the Ministry of Defence Animal Welfare Advisory Committee, and has served on a number of local Ethical Review Processes. He has served as a member of the Animal Procedures Committee, and was a founding member and Chair of its Housing and Husbandry Sub-Committee. He has recently edited the 8th edition of the UFAW Handbook on the Care and Management of Laboratory and other Research Animals.

## *c. Declarations of Possible Conflicts of Interest*

a. Previous contemporary employment of Mr Coulson (ASPI) and a senior member of Wickham staff between1995-99 at a contract research laboratory. During that period Mr Coulson had no authorities under ASPA 1986 and was employed as a Project Manager. The senior member of Wickham staff held a Personal Licence under the 1986 Act and was employed as an animal technician. There was no reporting or management relationship between these two posts. No perceived conflicts of interest arise through that prior association

b. Hospitality amounting to coffee and biscuits was offered by the PCD Holder at Wickham Laboratories and accepted by visiting ASPI Inspectors.