

# Training laboratory-housed non-human primates, part 2: Resources for developing and implementing training programmes

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**Keywords:** animal welfare; operant conditioning; positive reinforcement; refinement; socialisation; training

## Summary

Positive reinforcement training is increasingly being used to refine scientific, veterinary and husbandry procedures involving laboratory-housed non-human primates and to enhance primate care and wellbeing. However, there is a need and demand for greater access to resources designed to help laboratory personnel develop and implement appropriate training programmes. This paper includes a tabulated literature review of primate training, guidance on developing and implementing a training programme, and a detailed sample training protocol for training laboratory-housed primates to enter a transport container on request. The aim is to facilitate more comprehensive, systematic, humane and efficient use of training to refine primate use and management.

## Introduction

Part 1 of this paper presents the results, conclusions and recommendations of a survey to evaluate the extent to which non-human primates are trained in UK research and breeding establishments and to explore training knowledge and practice.<sup>1</sup> The survey indicates that:

- There is considerable scope for refinement of common scientific, veterinary and husbandry procedures through use of positive reinforcement training (PRT)
- Many constraints on training are perceived, rather than real, and can be overcome with information sharing and education
- There is a need and demand for greater access to resources designed to help laboratory personnel develop and implement appropriate training programmes.

This paper (Part 2) is designed to help establishments take positive action and implement the recommendations in Part 1 by providing:

1. A tabulated literature review of primate training
2. Guidance on developing and implementing a training programme, including resource and personnel requirements
3. A detailed sample training protocol for training laboratory-housed primates to enter a transport container on request.

It has been produced as part of a set of activities involving the authors and designed to facilitate more comprehensive, systematic, humane and efficient use of training:

- A continuing professional development course on training primates, coordinated by the University of Stirling ([h.m.buchanan-smith@stir.ac.uk](mailto:h.m.buchanan-smith@stir.ac.uk)) and the UK Institute of Animal Technology (see Heywood<sup>2</sup> for an informal review of a previous course on marmoset behaviour and welfare). The aims of the training course are to:
  - provide information on the costs and benefits of training the primate species most commonly used in laboratories
  - describe both the theoretical underpinning of learning theory and the practical application of socialisation, habituation and training of laboratory primates
  - demonstrate (video and live demonstration) and provide written details of tested protocols for training key tasks
  - discuss and provide practical solutions to primate management problems currently experienced by attendees in laboratories.
- PhD research funded by the Universities Federation for Animal Welfare Pharmaceutical Housing and Husbandry Steering Committee (UFAW PHHSC), which aims to identify ways to optimise the time investment involved with training, and to provide practical data on how PRT can be used in laboratories

- Production of audio-visual aids on interpreting primate behaviour and training primates for co-operation with common procedures, to be made available by the National Centre for the Replacement, Refinement and Reduction of Animals in Research (NC3Rs)
- Guidelines for refining food and fluid management in macaques being developed by a working group convened by the NC3Rs.

## 1. Literature on primate training

Table 1 shows reports in the scientific literature of primate training for co-operation with scientific, veterinary and husbandry procedures. Staff involved with primate breeding and use in research and testing should consult this table to identify opportunities for using training to refine such procedures (where possible, web links are given in the reference list). Staff can produce better science and reach their goals with better animal welfare by training primates to co-operate and investing in good relationships with them, rather than using forced restraint and coercion.<sup>1,3</sup> The table also includes examples of instances where training has been used to enhance animal care and well-being. Although, there is a long history of training primates to perform operant tasks for neuroscience and behavioural research, often using food or water restriction to motivate the animals, this work is not our focus and hence this literature is not included in Table 1.

## 2. Guidance on developing and implementing a training programme

Once the decision has been made to use training to refine a procedure, the next step is to develop and implement an appropriate training programme. This will typically involve:

- Selection of staff
- Educating staff about operant conditioning
- Developing animal training goals and strategies
- Implementing training sessions
- Record keeping and follow-up on progress.

With careful planning, adequate staffing and proper instruction, training can be integrated into existing practices and can be a rewarding endeavour for all involved.

### *Selection of staff*

The Biological Council<sup>4</sup>, Laule<sup>5</sup>, Colahan and Breder<sup>6</sup> and Young and Cipreste<sup>7</sup> detail some personnel requirements for successful animal training. In brief, competent trainers will have empathy with the animals to be trained, patience, a calm demeanour, be consistent in their behaviour and be able to analyse their own behaviour. Participants of the UK survey felt that a ‘team’ approach to animal training is the best way to ensure consistency and effectiveness, and that

the team should include animal technicians, scientists and veterinarians on equal and complementary terms (also see Rice et al.<sup>8</sup>). In addition, all staff should be aware of, and know the basics of, any training programmes in place, to ensure that they are interacting with the animals in a manner that is consistent with the detailed programme (e.g. not providing food rewards used during training out-with training sessions).

### *Educating staff about operant conditioning*

A staff member who is skilled and knowledgeable about operant conditioning<sup>1</sup> and training practices is likely to produce results more quickly than one who is not. For establishments where staff is unskilled in this regard, the time needed for educating personnel to use training methods effectively and the availability of such education must be considered. There are very few staff education courses on operant conditioning and training primates. We are aware of one other for primates housed in laboratories in addition to the aforementioned UK course. Dr. Steven Schapiro at the University of Texas MD Anderson Cancer Center (sschapiro@mdanderson.org) co-ordinates an 8-hour ‘Primate Enrichment and Training Workshop’ the main goals of which are:

- Presentation of the most recent advances in the application of environmental enrichment and PRT to the behavioural management of captive primates
- Provision of an essential complete guide to resources concerning environmental enrichment and training for primates
- Presentation of advanced techniques for solving difficult behavioural management problems using environmental enrichment and training techniques as tools.

For staff unable to attend the UK course or US workshop, a small number of audio-visual resources on animal training are available on international loan from the Shape of Enrichment ‘Enrichment and Training Video Library’ ([www.enrichment.org/collection.html](http://www.enrichment.org/collection.html)). Although primarily for use with zoo animals, these videos can be used for individual study or formal education.

There is also a growing body of literature available on how to train captive animals. Biological Council<sup>4</sup>, Laule<sup>5</sup>, Young and Cipreste<sup>7</sup> and Pyror<sup>9</sup> define and discuss training methods and related learning processes (e.g. operant conditioning, positive

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<sup>1</sup> The principle behind operant conditioning is that the consequence of a specific behaviour shapes the likelihood that the behaviour will happen again and hence the frequency with which it will occur. Positive reinforcement refers to a schedule of behaviour modification within the paradigm of operant conditioning whereby the frequency of a specific (required) behaviour is increased because something desirable is obtained on its performance (e.g. a favoured food).

**Table 1**

*Reports in the scientific literature of training of primates (often including habituation and desensitisation) for co-operation with scientific, veterinary and husbandry procedures and for enhancement of care and well-being*

Procedure	Species	Reference	Notes
Capture from the group/home cage	Rhesus, Bonnet and Long-tailed macaques	25	Using a transport container, singly-housed, juvenile females, combination of PRT and NRT, species differences were found in training effectiveness and corticosteroid response to confinement in the container after training
	Rhesus macaque	43,44 (video)	Using a transport container, young and adult males and females in a breeding group, combination of PRT and NRT, includes a training protocol
	Rhesus macaque	45	Using a transport container, adult males and females, singly- and pair-housed, combination of PRT and NRT
	Rhesus macaque	38	Using a transport container and chute, young and adult males and females, combination of PRT and NRT, includes analysis of time investment
	Rhesus macaque	46	File one at a time through a tunnel on cue, group-housed, combination of PRT and NRT, includes a training protocol
	Rhesus macaque	47	Using a modified transport container which is also used to restrain animals for behavioural testing, combination of PRT and NRT
	Stump-tailed macaque	48	Using a transport container, males and females
	Long-tailed macaque	49	Using a transport container, males and females
	Bonnet macaque	50	Using a transport container, includes a training protocol, young females, singly-housed
	Long-tailed macaque	36	Using a transport container, young females, singly-housed, combination of PRT and NRT
	Rhesus macaque, Baboon	51	Using a pole and collar system, adult males and females
	Drill	52	Diabetic adult male
	Chimpanzee	53	Using a transport container
	Squirrel monkey	54	Using a transport container
Transfer to a holding area	Japanese macaque	55	
	Chimpanzee	56	Indoors from outside, PRT, includes a training protocol and analysis of time investment
Handling	Primates	22	A review containing a table similar to this
Blood collection	Rhesus macaque	57	Using a restraint apparatus, adult females, group-housed, combination of PRT and NRT, includes a training protocol
	Rhesus macaque	58	Adult females, combination of PRT and NRT, includes a training protocol
	Rhesus macaque	59	Evidence of reduced cortisol response for trained animals
	Rhesus macaque	26	Adult males, pair- and singly-housed, combination of PRT and NRT, includes a training protocol
	Rhesus macaque	21	Young females, pair-housed, includes a training protocol
	Rhesus macaque	60	Adult females and male, combination of PRT and NRT, includes a training protocol, also faecal sampling
	Rhesus macaque	12	Combination of PRT and NRT, includes a training protocol, analysis of time investment and evidence of reduced cortisol response for trained animals
	Rhesus macaque	61	Adult males, pair-housed, via vascular access ports
	Long-tailed macaque	62	Diabetic animal trained to present a hand for finger puncture
	Stump-tailed macaque	63	Adult females, pair-housed, includes a training protocol
	Stump-tailed macaque	64	Males and females
	Vervet monkey	65	Adult females, singly-housed
	Drill	66	Adult male, clicker training, includes a training protocol
	Chimpanzee	20	Infant female, includes a training protocol
Chimpanzee	67	Young female, includes a training protocol	
Orang-utan	68		
Brown capuchin	69	Adult females, combination of PRT and NRT using restraint apparatus, includes a training protocol	
Primates	15	PRT, avoids use of squeeze-back, includes a training protocol, summarises techniques for training	
Blood and bile collection	Rhesus macaque	70	Males and females, animals are trained with PRT for wearing of jackets, to present subcutaneous access ports to the front of the cage, and to remain stationary

Blood pressure measurement	Rhesus macaque	71	Males and females
	Savannah baboon	72	Reduced cortisol response for trained animals
Injection	Savannah baboon	73	Adult males, singly-housed, PRT, includes a training protocol
	Rhesus macaque	10	Stand for intramuscular injection, combination of PRT and NRT, includes a training protocol
	Rhesus macaque	74	Adult females
	Rhesus macaque	75	Increased cortisol for non-trained animals
	Long-tailed macaque	62	Diabetic animal
	Lion-tailed macaque	76	
	Savannah baboon	77	Adult male, intramuscular injection in the arm, PRT, includes a training protocol
	Drill	52	Adult male, insulin injection, combination of PRT and NRT, includes a training protocol
	Chimpanzee	78	Subcutaneous injection into the abdomen, PRT
	Chimpanzee	79	Anaesthetic injection into the leg, PRT, trained animals showed lower mean values of total white blood cells, segmented neutrophils, glucose, cholesterol, and systolic and diastolic blood pressure, and higher hematocrit levels
	Chimpanzee	80	Anaesthetic injection, adult males and females, PRT, includes a training protocol and analysis of time investment
	Chimpanzee	81	Subcutaneous injection into the abdomen, PRT Intramuscular injection into the thigh, PRT or a combination of PRT and NRT
	Urine collection	Western lowland gorilla	82
Gibbons and Siamang Apes		17	Includes a training protocol
Apes		83	Includes a training protocol
Urine collection	Vervet monkey	84	Adult males, group-housed, PRT, includes a training protocol
	Chimpanzee	85	Adult females, single- and group-housed, PRT
	Chimpanzee	20	Infant female, PRT
	Chimpanzee	86	Adult females, PRT
	Gorilla	87	For collection on request, adult females, singly-housed, PRT, includes a training protocol
	Western lowland gorilla	88	Adult females, group-housed, PRT, includes a training protocol
	Common marmoset	89	For collection of first void, young and adult males and females, involves single-housing, includes a training protocol
	Common marmoset	11	For collection on request, includes a training protocol and analysis of time investment and effectiveness, PRT, does not involve single-housing
	Geoffroy's marmoset	90	Quantitative data for collection on request, PRT
	Emperor tamarin		
	Golden lion tamarin		
Cotton-top tamarin	91		
Pale-faced saki monkey	92		
Scent-mark	Common marmoset	93	Includes a training protocol
Saliva collection	Rhesus macaque	94	Adult males, using a pole or a screen method
	Western lowland gorilla	95 (video)	
	Western lowland gorilla	96	
	Common squirrel monkey	97	Chewing on a dental rope attached to a pole
	Common marmoset	98	Chewing on a cotton bud and allowing staff to retrieve the bud
Topical drug application	Stump-tailed macaque	99	Present heads through a Plexiglas plate, adult males and females, pair-housed
	Western lowland gorilla	100	Adult female, clicker training
Oral drug application	Long-tailed macaque	48	
	Pig-tailed macaque	101	Training to take juice from a syringe
	Savannah baboon	102	
Semen collection	Chimpanzee	103	Adult male, using an artificial vagina
	Chimpanzee	81	Adult males, using an artificial vagina, PRT
	Western lowland gorilla	104	Adult male
	Bonobo	105	Adult male
	Bonobo	106	Adult male
	Orang-utan	107	Adult male, using an artificial vagina
Insemination	Drill	108	Adult females, hindquarter presentation
	Bonobo	105	Adult female
	Bonobo	106	Adult female

Vaginal swabbing	Stump-tailed macaque Black-handed spider monkey	64 109	Adult females, group-housed, hindquarter presentation
Pinworm assessment	Chimpanzee	81	Presentation of the perineum for a tape test, PRT
Restraint	Common marmoset, Owl monkey	110	Tube restraint device
Eye tracking	Rhesus macaque Rhesus macaque	111 47	Using a primate chair, head restraint and eye coil Where neurophysiological recordings are not necessary, using an infra-red camera, transport container and face mask
Position emission tomography	Rhesus macaque	112	Using a head restraint device and primate chair
In home-cage behavioural testing	Rhesus macaque, Common marmoset Rhesus macaque, Common marmoset	113 47	Cognitive tests from the CANTAB, includes a training protocol
In home-cage weighing	Common marmoset	11	PRT, includes a training protocol and analysis of time investment and effectiveness
Stay at a particular location	Rhesus macaque	13	PRT, includes an analysis of time investment and effectiveness
Basic husbandry and veterinary examination	Western lowland gorilla Bonobo Primates  New World primates	114 (video) 115 6  18	Multi-task medical behaviours Describes in detail development of a programme of animal management based on training Describes in detail development of training programme for 17 species, evidence of species' differences in response to training
Cardiac evaluation	Bonobo	116	Physical evaluation, venepuncture and transthoracic echocardiography
Physical therapy for arthritis	Gorilla	117	Adult female
Reduce aggression during feeding	Chimpanzee Laboratory animals	23 5	Adult male, PRT Includes a training protocol, summarises techniques for training
Improve socialisation	Rhesus macaque Drill	118 108	
Management for reproduction	Drill, Western lowland gorilla	119	Reviews a range of PRT activities and benefits which could have application to captive breeding programs
Infant care	Western lowland gorilla Western lowland gorilla Western lowland gorilla White-cheeked gibbon Orang-utan Primates	120 121 122 17 123 6	Lists training goals for pregnant females and infants
Reduce abnormal and/or stereotypic behaviour	Drill Drill  Rhesus macaque  Olive hybrid baboons  Chimpanzee Orang-utan	14 124  125  126  127 128	Juvenile male, reduction in fence nibbling, self-biting, and examining and eating faeces and increase in positive social interaction Adult females, PRT, 4/6 animals trained to touch a target showed a reduction in stereotypic behaviour Juvenile males, reduction in whole-body stereotypic, cage- and self-directed behaviours, increase in activity levels Regurgitation and reingestion Female of a mated pair, PRT, reduction in stereotypic behaviour
Enhance enrichment programmes	Captive animals	129	Reviews use of PRT as an enrichment strategy to enhance psychological well-being

reinforcement, negative reinforcement, shaping or successive approximation, desensitisation, stationing, targeting, time out, regression). These resources serve as good introductions to the basic principles for training animals. Additionally, Pryor<sup>9</sup> and Young and Cipreste<sup>7</sup> give advice on choosing reinforcers, timing and size of reinforcers, schedules of reinforcement, shaping, establishing a cue, use of stimulus control, methods of using reinforcement to decrease unwanted behaviour, and clicker training. A number of further considerations for training may be obtained from the website of the Animal Behavior Management Committee of the American Association of Zoo Keepers: ([www.aazk.org/aazknew/committees/comm\\_animalbehavior.asp](http://www.aazk.org/aazknew/committees/comm_animalbehavior.asp)).

Briefly, the most important principles for implementing positive reinforcement successfully are to:

- Ensure that staff is well educated in the principles of training and appreciate that if training is not implemented correctly it may have a negative impact on animal behaviour (e.g. providing a primate with food after he/she has been aggressive will only serve to increase this undesirable behaviour)
- Choose an appropriate (i.e. biologically relevant and rewarding) form of positive reinforcement
- Establish clear rules to be followed for training (i.e. a clear and specific description of the behaviour that is to be reinforced)
- Ensure that access to the positive reinforcement that is to be used for training is restricted at other times (e.g. do not feed in the daily diet the food type used as a reward)
- Ensure that reinforcement occurs at the right time and place for the desired end result
- Initially arrange the situation to minimise the risk of failure (e.g. choose a location with minimal disturbance, but note that an isolated stressed animal is less likely to learn).

Another important step towards developing a training programme is to teach staff to be aware of their actions and how these affect primate behaviour.<sup>10</sup> It is also important that staff appreciate at the outset that the time investment they make in training animals is likely to be recouped later.<sup>11,12,13</sup>

### *Developing animal training goals and strategies*

An astute training programme will take into account the skill level of personnel, the research objectives, the natural history (behavioural ecology) of the species to be trained, and the temperament of individual animals. Failure to successfully train primates, or use of inappropriate techniques, may lead to frustration on the part of the animal (and staff) and a decrease in his/her well-being.<sup>14</sup> Therefore, it is recommended that a specialist in the field of behaviour modification be involved in the development of any training programme.

For a list of specialists willing to assist establishments in the development and implementation of behavioural modification programmes contact Dr. Mark Prescott ([mark.prescott@nc3rs.org.uk](mailto:mark.prescott@nc3rs.org.uk)).

The electronic discussion forum 'Laboratory Animal Refinement and Enrichment Forum (LAREF)' co-ordinated by Dr. Viktor Reinhardt at the Animal Welfare Institute ([www.awionline.org/Lab\\_animals/](http://www.awionline.org/Lab_animals/)) is another source of advice and contains regular correspondence on animal training, as does the Animal Training List Serve of the American Association of Zoos and Aquariums (contact [mthompson@lifegate.net](mailto:mthompson@lifegate.net)). It may also be possible to contact the authors of the papers listed in Table 1 to take account of their experiences in implementing training and addressing any problems. For example, Laule et al.<sup>15</sup> and Wolfensohn and Honess<sup>10</sup> give advice for laboratories that are moving toward an animal management system based on training with positive reinforcement, and Whittaker et al.<sup>16</sup> describe development of a behavioural management programme for chimpanzees at the MD Anderson Cancer Center Science Park.

Many North American zoos have extensive training programmes for veterinary management and breeding of their primates and, as such, can be valuable sources of advice for research and breeding establishments. Colahan and Breder<sup>6</sup> describe the philosophy and framework for a training programme at Disney's Animal Kingdom (DAK), emphasising setting goals, planning, implementing, documenting, evaluating and re-adjusting. Further information on the cues and criteria used for husbandry training of gibbons and siamangs at DAK is given in Richards et al.<sup>17</sup> and Savastano et al.<sup>18</sup> who describe the development of a training programme for seventeen species of New World monkeys at the Bronx Zoo involving a wide range of basic and advanced behaviours (e.g. syringe feeding, targeting, scale and crate training, and transponder reading). The authors include details of the cues used, criteria for successful learning, time scales for learning, logistical and animal challenges and solutions.

Before any training programme is started it is essential to put in place some key elements which will facilitate the smooth running of the programme and produce the highest success rate. Time should be allocated for staff involved in the programme to meet to plan exactly how the programme should be run, who has responsibility for what, which behaviours will be reinforced, and which cues and rewards will be used.

It is useful if one person has overall responsibility for the training programme to ensure a consistent approach, although a number of people can be involved with the training. Consistency of technique between trainers is important; otherwise, for example, the

wrong behaviours can be accidentally reinforced and training disrupted. Therefore, the method and sequence of training should be agreed by all involved at the outset and written protocols should be established which clarify approved and, where appropriate, non-approved, training methods. Laule<sup>5</sup> includes a sample protocol for training for co-operative feeding, and a detailed sample protocol for training laboratory-housed primates to enter a transport container on request is included below (see also Table 1).

Station (approach a specific location) and target (touch a specific object) are useful behaviours to train in the early stages of a training programme because they can be used in a number of circumstances. For example, a primate that learns to follow a target can be requested to sit on a weighing scale in the home cage, thereby avoiding capture, restraint and removal from the cage.<sup>11</sup> These behaviours are especially helpful for managing primates that are not easily handled, display aggressive behaviours, or live in large enclosures or groups.

Training is easiest with animals that are well socialised to humans<sup>11,19,20</sup> and it is recommended that, as a first step, all animals are habituated to accept food from the hands of staff. This can be achieved initially by placing food in front of the animals' enclosure within reach of the animals and stepping back from the enclosure front, gradually decreasing the distance between human and animals over time until food is accepted from the hand. (Eye contact should be gradually increased for marmosets, but not with macaques, who should not be looked at directly). Alternatively, food can be provided on the end of a pole that can be used to control the distance between animal and human (see sample training protocol below).

In order to successfully train primates it is essential that all animals involved can be individually and easily recognised. If individuals are not naturally distinguishable, this can be achieved, for example, by using collars and tags, site-specific hair clipping (e.g. from the tail) or subtle coloured dye marks on the fur. All individuals should be named, preferably not with names which reflect less amenable aspects of their personality (e.g. Grumpy or Biter) but with those that are distinctive and easily spoken (e.g. Angus or Erin).

It is important to recognise that not all animals may be able to be trained.<sup>13,21,22,23,24</sup> Primates have distinct personalities and aptitudes and, as such, individuals are bound to vary in both their willingness and ability to learn. There may also be sex, species and group dynamic differences that affect behaviour and trainability.<sup>6,18,24,25,26,27,28,29</sup> Trainers, therefore, are required to be knowledgeable about these aspects and plan their protocols accordingly.<sup>6</sup>

Avoid separating primates for training as this may cause stress and the animals will learn less well. Having said that, it is important to understand and work with the dominance hierarchy, training the most dominant animal first. Be aware that if a dominant animal is nearby, watching a subordinate being trained, the subordinate can become unsettled, unfocussed and may be reluctant to accept the reward (K. Morris, pers. comm. to H.M. Buchanan-Smith).

### *Implementing training sessions*

For some facilities the principle cost of training is the time investment involved in implementing the behavioural modification process.<sup>1</sup> In order to facilitate efficient use of time, facilities can keep training sessions short (e.g. 2-10 minutes). Training sessions should be short anyway, so as to maintain the animal's interest, but the optimum length varies between species and individuals.<sup>18</sup> Although more training sessions in a given time enables training goals to be achieved faster, training can still be achieved with three to four sessions per week. Once the desired behaviour has been well established, reinforcement for performing the behaviour can be even less frequent (e.g. once every two weeks), but it is important that the behaviour does continue to be reinforced or else the animal will stop performing the desired response and will require re-training.

Establishments can take small steps towards a proactive behavioural management programme by incorporating training into the daily routine (e.g. whenever primates are being fed). Sessions can be relatively passive depending on the desired behaviour. For example, training primates to initially go into a transport container may be as simple as placing their food closer to the transport container over a period of days, and eventually inside it. Facilities with young animals can begin by exposing those animals to unfamiliar objects that they may encounter later in life (e.g. stethoscope, syringe, primate chair). Curiosity towards novel objects in young primates has been noted by many authors, whereas adults, in comparison, may be considered conservative due to the accumulation of experience.<sup>30,31,32,33,34,35</sup> Associating unfamiliar objects with feeding times can help desensitise the animals to these objects.

Although training can be easily incorporated into the daily routine, the time invested to develop and implement a structured programme can have tremendous benefits. Eventually, procedures can be performed quickly, efficiently and calmly, saving time in the long run.<sup>11,12,22,36,37,38,39</sup> Therefore, instead of viewing training as an additional luxury to include in daily operations, managers should consider training as essential to the successful operation of an animal facility and plan and budget accordingly. For establishments with large numbers of animals, it may

help to prioritise and focus on key studies, animals, behaviours or procedures, (e.g. those that cause most suffering, for which training protocols exist, or for which co-operation can be achieved most quickly).

Training methods should be based on positive reinforcement since this is considered to be the most humane method of training.<sup>5,9,15</sup> Negative reinforcement should only be used when positive alternatives have been exhausted. If negative reinforcement must be used (e.g. for training aversive procedures), it should be used in combination with positive reinforcement.<sup>40</sup>

### Record keeping and follow-up on progress

Records of training performance should be kept for each session and regularly reviewed (e.g. weekly) for each animal, noting their achievements and any abnormal, unpredictable or otherwise significant behavioural irregularities at each training session. Such records can be used to identify when to begin to train the next behaviour/task in a programme of successive approximation, to help monitor success or otherwise of the training programme, and to justify to management the time investment involved.<sup>41</sup> Knowing when to train the next step of successive approximation in the programme is important; the primate must be allowed to succeed and be reinforced but, in order to maintain interest in the task and to ensure progress to the final desired behaviour, new goals must be introduced. If the primate has not succeeded to obtain many rewards during the session, reverting to earlier learned behaviours at the end of the session is important so that the session ends on a positive note.

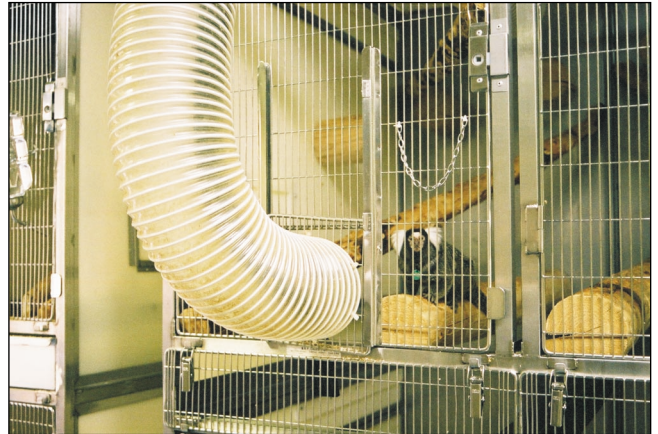
### 3. Sample training protocol: Training laboratory-housed primates to enter a transport container on request

Use of a transport container (box) enables primates to be confined and moved from one cage or enclosure to another without chemical restraint or anaesthesia. This is useful for:

- Veterinary management (e.g. health examination, collecting physiological samples, administering topical or injectable medications)
- Breeding management (e.g. setting up temporary pairings)
- Husbandry (e.g. relocating animals during cage cleaning or maintenance, weighing)
- Enrichment (e.g. relocating animals to remote play or exercise areas)
- Research purposes (e.g. temporary move to a metabolism cage for collection of faecal samples or to a testing site for cognitive studies).

A protocol for training laboratory-housed primates to enter a transport container on request is given below. The protocol was developed for common marmosets at

the MRC Human Reproductive Sciences Unit in Edinburgh, where it has been used successfully. The protocol can be modified for use with other species; some suggestions for macaques are given here. An alternative for moving marmosets between cages is the use of plastic tubing (Figure 1).



**Figure 1.** Marmosets will readily travel through air ventilation ducting, which is cheap, flexible and easily attached to enclosures. It can be used to move marmosets between locations, obviating the need for capture, and marmosets will readily travel long distances (> 10m) once they are familiar with the ducting. If long vertical stretches are included, it is recommended that some strips of matting are inserted into the ducting (as seen in this photograph) to allow the marmoset to grip.

**Goal:** The goal is to train laboratory-housed primates using PRT to enter a transport container (box) on request, and to remain relaxed whilst they are shut in and transported to another location.

**Select a training reward:** A 'training reward' should be agreed upon which is a favoured food that will only be used during training sessions and will not be part of the regular diet (e.g. raisins, cornflakes, dates or marshmallow for marmosets. Although high sugar/high fat foods should be avoided, marmosets work more reliably for marshmallow than for other rewards). Rewards should be prepared prior to the start of each session, so there is no delay in producing a reward once training has begun. Each reward should be small; no more than one mouthful (i.e. 1/2 raisin or 1/12<sup>th</sup> small marshmallow for marmosets). Problems of aggression can occur if the reward is too highly favoured. Such problems can be overcome by using a slightly less favoured food with occasional 'jackpots' of the most favoured food and by giving an individual a reward only in his/her 'reward zone' (see below).

**Select bridges and cue words:** Selection of secondary reinforcers (bridges) and cue words should be established when setting up a training programme. The bridge is a sound or word which is used to mark the desired behaviour (a sound is usually more consistent if many trainers will be working with the animals). It is



used to say to the animal 'what you are doing right now is the thing you are getting a reward for'. The bridge enables the trainer to reward the animal for doing something when he/she is physically distant from the trainer (i.e. not at the front of the cage). Commercially available clickers can be used for macaques, but these are often too loud for more timid primate species (e.g. marmosets), so a vocal tongue click can be used or the click of a retractable pen. Advice on establishing the bridge is provided below. Cue words are used to provide the trainer with control over which animal performs what behaviour when; similar to when we ask a dog to sit. In this protocol it may be decided to use the primate's name followed by the word 'box' to request that he/she goes into the box (e.g. 'Angus, box!'). A calm but upbeat voice should be used to encourage a response. If the behaviour is performed readily (e.g. target), then the cue word can be introduced early. For complex behaviours that require to be shaped (training to urinate on request), it is best to increase the frequency of the behaviour first, before introducing the cue word.

**Define staff responsibilities:** Whilst a number of people can be involved in training the animals it is best if each person has responsibility for a particular group or room, and that they remain the sole trainer of that group/room until the behaviour is established. This ensures the approach is consistent throughout the early stages of establishing a behaviour. However it may be useful to have two or more people working with larger groups of animals to prevent squabbles over access to the food, and to facilitate cover over holidays and sick days. If there are multiple trainers, each person should take responsibility for certain animals and only reward those animals. Each trainer should ignore animals that they are not responsible for training. The primates will soon learn who their dedicated trainer is and ignore the other trainer.

**Establish hand feeding:** The first behaviour it is essential to establish is hand feeding, whereby the animals will come forward and take a food reward from the trainer. Even at this early stage it is useful to establish 'reward zones' whereby each individual animal in the group only gets a food reward whilst in a particular location (e.g. top, left-hand side of the cage). If each individual has his/her own location, this will help to prevent squabbling and food stealing. Nervous or hesitant individuals who are reluctant to come and take food from the trainer can be offered food on a pole inside the cage. The distance between the trainer and the food reward can then be decreased gradually until the animal will take the food directly from the trainer. The distance should only be reduced once the animal is reliably taking the reward at the longer distance (i.e. takes the reward nine times out of ten). If the animal fails to take the reward at the shorter distance for three consecutive occasions, the distance should be increased to one where he/she is performing reliably

before being shortened again. Any undesirable behaviour (e.g. aggression between conspecifics, threats or aggression towards the trainer, abnormal behaviours such as fur-plucking) should be ignored and a time-out introduced. The time-out must occur immediately after the undesirable behaviour starts and involves the trainer turning round so they have their back to the cage, and food rewards are out of sight. Training can resume only once the animals are calm and relaxed again. It is essential that the animal is never rewarded for performing an undesirable behaviour and the trainer must be particularly aware of their own behaviour in regards to this. It is particularly easy to train primates to grab and scratch the trainer when giving them their food reward. However, it is important not to withhold the reward from snatchers if they have performed the desired behaviour. Rather give a double reward when they take the reward calmly. Hand feeding can be performed two to three times per day in short sessions (~2 minutes) until all animals in the group will eat in this way.

**Establish the bridge:** Once all animals in the group can be hand-fed in a relatively calm manner (some excitement is acceptable, as enthusiasm for getting their rewards will encourage the animals to work and maintain interest during training sessions) the bridge can be established. To do this the click (or whatever bridge has been decided upon) should be paired with the arrival of the food reward, so the trainer should click immediately before (i.e. <1 second) the food is delivered to the animal's mouth. The animal may be slightly startled at first but careful repetition will soon enable him/her to associate the click with the food. The trainer should then click just before the food reward is delivered and if the animal has made the connection between the click and the food reward he/she will look for the food reward. If this does not happen the training has been too fast and the trainer should go back a stage to allow the animal to make this connection. It is vital that this stage is not rushed (e.g. minimum of three to four sessions) as a strong bridge is essential for all the later stages of training. The aim of this part of the training is to establish a response whereby the animal will wait attentively for his/her food reward for 2-3 seconds following the click. Again, short sessions are preferable (e.g. maximum of 8 minutes) or until the animal earns eight rewards (but this may vary depending upon the size of group). If time allows, there can be multiple sessions each day, and training will be quicker but, if time does not permit, transport box training can also be achieved with three to four sessions per week.

**Begin target training:** It is then useful to train the animals to hold a 'target'. Target training enables the trainer to keep stationary one or more animals whilst working with another, and also enables the trainer to guide the target animal into the transport box once it is

introduced. The target should be an easily replaced, easily cleaned object which the animals do not see out of training sessions (a pen would not be suitable). Each animal in the group should have its own target which is identifiable by colour<sup>2</sup>. A suitable target for marmosets is a plastic teaspoon; black and white are the best colour combination for pairs. In the individual's reward zone the trainer should hold a food reward behind the target and click the animal as he/she touches the target whilst reaching for the reward. This should be repeated until the animal deliberately touches the target on five consecutive occasions it is offered. Next the animal should be clicked and rewarded only if he/she actually holds the target, however briefly (the click must come whilst the animal is holding the target). Once the animal will hold the target on five successive occasions the duration of hold should be increased, first 1 second for five consecutive occasions, then increasing to 3 seconds, 5 seconds and 10 seconds, each for five successive occasions. (Actually, with marmosets, holding is achieved relatively quickly and successive steps may not be needed for all individuals). Longer durations can be introduced if required. This is done by clicking only after the desired duration of hold has passed. If, at any point, the animal fails to hold the target for the required duration on two consecutive occasions, or three out of ten presentations, the trainer should go back to the last stage and re-establish that. It is also often useful to start a session at a slightly lower level than previously achieved as this allows the animal to have easy success and helps maintain their interest and enthusiasm. For this reason it is also important to finish every session with a short run of successes, even if this means working at a lower level. Sometimes the primates will become a little overenthusiastic and try to hold their cage-mate's target. This behaviour should be ignored, and not rewarded either by food or a vocal response.

**Move the target:** The target should now be moved around the cage and the animal rewarded for holding it wherever it is presented. The target should gradually be moved away from the reward zone, again going back a step if the response becomes unreliable. If desired, a verbal cue can now be introduced with the required behaviour. As the target is presented, the trainer should use the cue word (e.g. 'Angus, hold!'). The cue word should be used on every occasion the target is presented.

**Introduce the transport box:** To introduce the transport box, it should be carefully and securely placed on the front of the cage with the box door open and the

animals given access to it (Figure 2). They should be allowed a few minutes to explore the box if they wish to. At first, for social reassurance, several animals can be target trained in the box simultaneously. However, if there is a requirement to separate just one individual, the trainer should then ensure all but one of the animals in the group is holding their targets away from the box (in groups of more than two primates it may be useful to have a second trainer to target the other primates, whilst the original trainer works on box training). The trainer should then place the target of the animal he/she is working with at the end of the transport box. The animal may then enter the box. If there is hesitancy the target can be placed near the box door as a guide for the required behaviour, and the animal rewarded for holding it. The target can then be gradually placed further inside the box, each time clicking and rewarding the animal for holding it until he/she will come into the box in a relaxed manner (Figure 3). The duration of wait in the box should then be increased as described for target training until the animal will remain in the box for 10-20 seconds.

**Habituate the animal to the box door opening and closing:** Next, the door needs to be shut. This creates noise and some movement in the box, which may cause the animal to become unsettled. This can be reduced by ensuring the door is running freely prior to training, but, providing the target response has been well established, the animal should remain holding the target (Figure 4). The trainer can click the continued hold at first, providing he/she does so only if the animal remains relatively calm. The animal should be requested to hold the target and the door closed and immediately opened. The animal should remain in the box once the door has reopened for at least 2 seconds, then clicked and rewarded; this is to ensure that he/she does not learn to rush out of the box as soon as it opens. The animal should then be asked to leave the box using their target. This process can be repeated until the animal becomes habituated to the door opening and closing (i.e. no longer takes any notice of it).

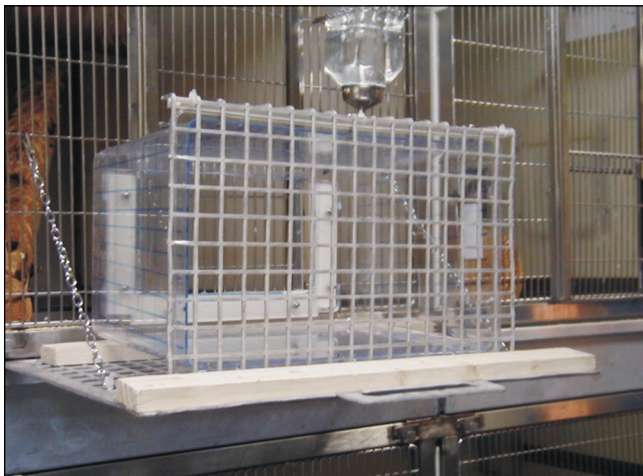
**Increase the duration of time the animal spends in the box:** The duration of time the animal spends in the box can then be increased by leaving the door closed for 2 seconds, then 5 seconds, working up in small increments until the box remains shut for 5 minutes. The animal does not need to hold his/her target for this entire time, but he/she should always be recalled to hold before the door is reopened to control his/her exit. The animal should be clicked and rewarded for calm behaviour in the box during these waits. Bear in mind that some primates, especially younger animals, may become bored during these waits. The trainer can ask for intermittent target holds during this time to prevent undesirable behaviours occurring due to boredom. The duration of wait in the box should be varied from no wait

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<sup>2</sup> Note that most diurnal New World primates have polymorphic colour vision – only heterozygous females can be trichromatic (similar to most humans), whereas homozygous females and all males are dichromatic (colloquially colour blind).<sup>42</sup>

to the maximum being trained at that time to keep the animal's concentration and interest. Cue words can now be introduced as described above, so that the behaviour can be requested verbally. Given that the same task of holding the target is required, no new cue words need to be introduced (i.e. use 'Angus, hold!' as before).

**Habituate the animal to transport in the box:** The box can now be carefully detached from the home cage and placed a short distance away, ensuring visual, olfactory and auditory communication with conspecifics is maintained. The animal should be held in the box for 1 minute, clicked and rewarded for remaining calm, before the box is returned to the home cage and the animal released (following a target hold whilst the door is opened). This should be repeated and providing the response remains reliable (i.e. nine out of ten requests) the distance between the home cage and the box increased until the box is out of visual contact, then out of auditory and olfactory contact. Once this is reliably established longer duration waits can be introduced, and releases at different environments (e.g. play areas).



**Figure 2.** Transport box is carefully attached to the home cage.



**Figure 3.** Marmoset calmly holding the target in the transport box.



**Figure 4.** Marmoset calmly accepting a food reward while the door of the box is shut.

Providing the routine is kept varied, and desirable behaviours continue to be reinforced, the trained primates should now be easy to catch and move to new locations. The bridge can be used in other situations to reinforce desired behaviours and target training can be used, for example, to select animals for visual inspections or applying topical medications whilst the animal remains in his/her home cage.

## Acknowledgements

Special thanks to Keith Morris and staff at the MRC Human Reproductive Sciences Unit for their continued support and encouragement. We appreciate discussions with Jean McKinley and staff at Dstl Porton Down on training protocols. MJP is employed by the NC3Rs. HMB-S was supported by a grant from the European Commission (QLRT-2001-00028). VAB was supported by the UFAW PHHSC.

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