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Title of Project: Minimally Invasive Deep Brain Stimulation

Summary:

Brain machine interface (BMI) (or brain computer interface (BCI)) devices currently require invasive surgery to be implanted. As miniaturisation and wireless technologies continue to advance, there are an increasing number of opportunities to employ BMIs as therapeutic agents to treat neuropsychiatric conditions. However, safer implantation methods are needed in order to avoid the complications associated with conventional neurosurgery and allow 'high-risk' surgical patients to access these devices. Minimally invasive approaches, such as catheterisation techniques used to deploy coronary artery stents, would be an attractive alternative to craniotomies *if* devices positioned in the vasculature could exert the same effects achieved by BMI devices sited in the cerebral parenchyma. Placing devices in the cerebral vasculature to record EEG has been proposed by Penn *et al* '73, and for stimulation by Llinás *et al* '05 and Teplitzky *et al* '14. Recently, the Neural Bionics Laboratory at The Royal Melbourne Hospital designed such a transvascular BMI device for recording purposes. The device was a stent-electrode array and they used it to record brain signals in the Ovine large animal model (data under review for publication). However, its capacity to stimulate across the vasculature, a promising concept, had not been assessed and thus provided the basis for the work reported here.

At the outset it was deemed that the Ovine model was an appropriate animal in which to study the device. The first reason for this was that a target for stimulation, the motor cortex, lay adjacent to the superior sagittal sinus. This sinus was in fact the cerebral vessel that the stent-electrode array had previously been positioned in for recording purposes. The second reason to use sheep was that the catheter deployment technique used for the devices had already been developed for this animal; thus, significant time could be saved continuing to use this model as opposed to optimising it for another animal model.

The hypothesis posed was that: ***a stent-electrode array positioned in the superior sagittal sinus can transvascularly stimulate the Ovine motor cortex***. Proving this hypothesis would lay the groundwork for a paradigm shift towards transvascular stimulation as opposed to intra-parenchymal cerebral stimulation.

In total, nine sheep were used for the stimulation studies (only one was excluded secondary to debris blocking the device's external plug). The devices were stimulated at a range of different time points post implantation. This was done in an attempt to account for endothelialisation that was predicted to take place about the stent-electrode arrays. In addition a range of stimulation parameters and electrode configurations were trialled when stimulating the Ovine motor cortex. Across the

experiments a range of motor activities were induced. These included sustained contractions localised to the neck, fasciculations about the supra-lip region, and flank fasciculations. In addition, on an occasion when the stimulation equipment malfunctioned and a fuse blown, a 50 second generalised seizure was induced in one sheep.

However, while a variety of contractions were induced, it was not a certainty that these represented actual stimulation of the Ovine motor cortex instead of muscular activity induced by current spread. Indeed, it became apparent during post-mortems that some of the cables to the stent-electrode arrays had been severed (this was not the case in devices that had been implanted for less than one month). In such animals the neck contractions had been at the cable exit site from the jugular vein. Furthermore, a number of contractions were induced in the setting of unipolar electrode configurations. That is, an electrode on the stent-electrode array served as one pole, and a ground electrode placed subcutaneously on the flank or skull acted as a return. Using these configurations with a flank return electrode, the flank contractions were noted.

Thus while the work to date has attempted to test and prove the lead hypothesis, more work needs to be done. At this stage, work continues in the setting of an established stent-electrode array device and method for deploying it in sheep. In addition, stimulation equipment that is compatible with the stent-electrode array, and capable of driving a current through it, is in place. Furthermore, an animal handling setup consisting of a Corriedale specific sling and head collar has been designed, and video recording equipment, with LED trigger lights and accelerometers to document movement have been fabricated and are working reliably. Finally, a range of stimulation configurations and parameters has been trialled at different time points. Future work will focus on bipolar stimulation in order to avoid current spread phenomena, and at higher current strengths.

Hypothesis vs Findings

Hypothesis:

A stent-electrode array positioned in the superior sagittal sinus can transvascularly stimulate the Ovine motor cortex.

Findings:

Stimulation paradigm: An AMPI Master-9 and ISO-Flex stimulus isolator were used to deliver current-controlled impulses to the stent-electrode arrays. The frequencies explored included those of 20 Hz, 50 Hz, and pulse trains. The pulse trains consisted of 200 μs monophasic pulses at 2,500 Hz for a total of 5 pulses, and these trains were conducted at a frequency of 1 Hz for 5-20 seconds. When stimulations of 20 Hz or 50 Hz were used, monophasic pulses of either 200 μs or 300 μs were used. When pulses were of 200 μs duration the maximum current used was 6 mA whereas when the pulses were 300 μs the maximum current used was 3 mA. Distant return electrodes were used for monopolar studies, and consisted of either a stainless steel disc donut subcutaneously deployed on the dorsal aspect of the sheep's posterior, termed the rear return, or a platinum sheet positioned subcutaneously of the sheep's skull, termed the head return. In addition to monopolar studies, bipolar studies employing a range of configurations were also trialled. When it was desirable to investigate current strengths at 10 mA, two electrodes of the stent-electrode array were combined at the lead connection interface outside of the sheep. Stimulation studies were conducted over a range of dates post-implantation and a sheep was never stimulated for more than four hours.

1.1 Transvascular stimulation studies

1.1.1 B50

1.1.1.1 Experiment #1

B50 was stimulated whilst the animal was standing in its usual metabolism cage. It was in a long, well-lit room in the company of ten other sheep (each in their respective cages) to promote an environment in which the sheep felt safe. There were no signs of distress such as stomping or an increased work of breathing, it accepted food from the examiner's hand, and it appeared that the animal tolerated the stimulations studies well.

For the experiment no tethering equipment was used and motor activity was monitored by observation and video recording alone. The stimulation configuration available was restricted to monopolar stimulation, where the monopole consisted of three shorted electrodes, namely electrode 2, 4, and 6, conducting current to a distant return, in this instance the rear return. The parameters trialled included a frequency of 20 Hz stimulating for 2 seconds with a biphasic pulse, led by a cathodal current, with a 200 μs phase duration.

While there was a behavioural response to stimulation, characterised by the sheep lowering its head, with a threshold of 2.2 mA, there were no other discernable contractions.

1.1.1.2 Experiment #2 – **A seizure**

Four days after the first experiment on B50 a second experiment was done to replicate the behavioural response and study the effects of different stimulation parameters. The setup for the animal was the same and the stimulation parameters were initially the same. However, the behavioural response as initially seen was not observed. There were three connections on the plug to the stent-electrode array that could be used to direct current through the three shorted electrodes. When the connection was changed, even though the AMPI Master-9 was powered off, the stimulus isolators delivered an unexpected current. After the unknown current was sent through the device and a 50 second generalised seizure was caused. The sheep did not pass urine or faeces, and no physical damage was sustained from its fall. It was standing shortly after seizure termination and ate food from the examiner's hands with no difficulty.

Subsequent to the event, the A.M.P.I. Master-9 and the two ISO-Flex stimulus isolators were all tested. The A.M.P.I. Master-9 was operating normally, however one ISO-Flex stimulus isolator was discharging a constant current and voltage, while the second isolator was not sending out any charge, even when the A.M.P.I. Master-9 was directing it to. The second isolator was found to have a blown fuse. The fuse was designed to break when a current of 80 mA passed through its wire. When a working fuse was introduced, this stimulator then began to discharge in an uncontrolled manner, as was the case with the first stimulus isolator. A key difference between the isolators was that the second had had its internal resistor decreased from 33.2 kΩ to 12 kΩ in order to allow that stimulator to pass a current up to 20 mA instead of the standard 10 mA. Both devices were returned to the manufacturer for further diagnostics and repair. The transistors were found to be damaged and the 12 kΩ resistor changed back to a 33 kΩ resistor, which was seen by the engineering team as the cause of the malfunction.

Given the malfunction with a biphasic setup, biphasic stimulations were abandoned for the remaining experiments.

1.1.1.3 Experiment #3

In order to explore the effects of additional stimulation parameters a third study was performed with B50.

The stimulation configuration available remained restricted to monopolar stimulation, where the monopole consisted of three shorted electrodes, namely electrode 2, 4, and 6, conducting current to a distant return, in this instance the rear return. The parameters trialled included a frequency of 60 Hz stimulating for 1-15 seconds with a monophasic pulse, led by an anodal current, with a 300 μs phase duration.

While there was a behavioural response to stimulation, characterised by the sheep lowering its head, with a threshold of 2.7 mA, there were no other discernable contractions.

1.1.1.4 Experiment #4

In order to explore the effects of additional stimulation parameters a fourth and final study was performed with B50.

The stimulation configuration available remained restricted to monopolar stimulation, where the monopole consisted of three shorted electrodes, namely electrode 2, 4, and 6, conducting current to a distant return, in this instance the rear return. The parameters trialled included a frequency of 50 Hz stimulating for 2 seconds with a monophasic pulse, led by an anodal current, with a 500 μs phase duration.

While there was a behavioural response to stimulation, characterised by the sheep lowering its head, with a threshold of 2.5 mA, there were no other discernable contractions.

1.1.2 B55

1.1.2.1 Experiment #1

B55 was stimulated whilst the animal was standing in its usual metabolism cage. It was in a long, well-lit room in the company of ten other sheep (each in their respective cages) to promote an environment in which the sheep felt safe. On inspection of animal's neck, where the electrode wires were connected to a plug, the exit site had become dehisced with discharge seeping from the opening. In spite of this, the animal appeared in relatively good condition. Additional antibiotics were administered.

For the experiment no tethering equipment was used and motor activity was monitored by observation and video recording alone. The engineering team indicated that five of the eight electrodes were working, which included electrodes #1, #4, #5, #7 and #8. Monopolar stimulation alone was studied with the rear ground used as the return. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse, either as an anodal or cathodal current, with a 200 μs or 500 μs phase duration. Currents used were either 3 or 6 mA with phases that were 200 μs in duration or 1, 2, or 3 mA with phases that were 500 μs in duration.

There were no responses to any stimulation parameters trialled.

1.1.2.2 Experiment #2

B55 was subsequently stimulated using a sling setup in order to relax the antigravity musculature of the legs. The animal's neck, where the electrode wires were connected to a plug, remained in a dehisced state. Tethering, accelerometer, and electromyography setups were in place.

Five electrodes, #1, #4, #5, #7 and #8 were studied as per prior advice from the engineering team. Monopolar stimulation alone was studied with the rear ground used as the return. The parameters trialled were as per the first experiment in this animal.

There were no responses to any stimulation parameters trialled.

1.1.3 B61

1.1.3.1 Experiment #1

B61 was stimulated whilst the animal was standing in its usual metabolism cage. It was in a long, well-lit room in the company of other sheep to promote an environment in which the sheep felt safe. There were no signs of distress such as stomping or an increased work of breathing, and it appeared that the animal tolerated the stimulations studies well.

For the experiment no tethering equipment was used and motor activity was monitored by observation and video recording alone. The stimulation configuration available was restricted to monopolar stimulation, using any one of the seven electrodes in combination with the rear ground as the return. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse, either as an anodal or cathodal current, with a 200 μs or 500 μs phase duration. Currents used were either 3 or 6 mA with phases that were 200 μs in duration or 1, 2, or 3 mA with phases that were 500 μs in duration.

When the stimuli were of 500 μs phase width, no responses were seen when a current strength of 1 or 2 mA was used. However, when 3 mA current strength was used, and a frequency of 50 Hz with an anodal phase, electrodes 2-8 were associated with fasciculations around the flank region where the rear return was located; which in this configuration acted as the cathode. When these electrodes acted as the cathode, there were contractions noted about the left foreleg. When the frequency was reduced to 20 Hz, these responses were only noted with electrodes 3 and 4.

When the stimuli were of 200 μs phase width, no responses were seen at 3 mA, however at 6 mA some responses were seen. At 20 Hz with the electrodes 1-8 acting as the anode and the rear return as the cathode fasciculations as were evident with 500 μs phase widths were again observed. However, when the electrodes acted as a cathode, no responses were seen. When the stimulation frequency was increased to 50 Hz, no responses were seen.

1.1.3.2 Experiment #2

B61 was again stimulated using a sling setup in order to study addition stimulation configurations. There were no signs of distress, and it appeared that the animal tolerated the stimulations studies well.

The stimulation configurations included bipolar orientations as well as monopolar orientations with either the head return or rear return used. Each of the seven electrodes available was used. The parameters trialled included a frequency of 20 Hz for 3 seconds with a monophasic pulse, either as an anodal or cathodal current, with a 200 μs phase duration. Currents used were either 3 or 6 mA. In addition, electrodes were joined as doublets in order to increase the effective surface area and stimulations performed in a bipolar manner, doublet to doublet, at a current strength of 10 mA with 200 μs phase widths at 20 Hz for 3 seconds.

When the head return was used there were no responses to any of the stimulations. However, when the rear return was used with the same stimulation run, flank fasciculations were evident when the rear return acted as the cathode. This was current dependent with no response noted at 3 mA, but responses elicited at 6 mA. In contrast to the first experiment with this animal, monopolar stimulation to the rear return with the electrodes of the stent-electrode array acting as the cathode was associated with no response.

Bipolar configurations comprised of electrode 1 connected to 2, then 2 to 3, then 3 to 4 and so on, were associated with no response. Doublet configurations in a bipolar arrangement, with electrode 1 joined to 2 and this doublet connected to a doublet of electrode 3 joined to 4, at 10 mA, was not associated with any response either.

1.1.4 B62

1.1.4.1 Experiment #1

B62, a sheep with its device exiting the right jugular, was stimulated whilst the animal was standing in its usual metabolism cage. It was in a long, well-lit room in the company of other sheep to promote an environment in which the sheep felt safe. There were no signs of distress such as stomping or an increased work of breathing, and it appeared that the animal tolerated the stimulations studies well.

For the experiment no tethering equipment was used and motor activity was monitored by observation and video recording alone. The stimulation configuration available was restricted to monopolar stimulation, using any one of the eight electrodes in combination with the rear ground as the return. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse, anodal current alone, with a 200 μs or 500 μs phase duration. Currents used were either 3 or 6 mA with phases that were 200 μs in duration or 1, 2, or 3 mA with phases that were 500 μs in duration.

Using these anodal currents, the first electrode alone was associated with a contraction of the right neck. All other electrodes did not produce a response.

1.1.4.2 Experiment #2

B62 was again stimulated using a sling setup to relax the anti-gravity muscles in its limbs and in order to study addition stimulation configurations. There were no signs of distress, and it appeared that the animal tolerated the stimulations studies well.

The stimulation configuration available was restricted to monopolar stimulation, using any one of the seven electrodes in combination with the rear ground as the return. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse, either as an anodal or cathodal current, with a 200 μs or 500 μs phase duration. Currents used were either 3 or 6 mA with phases that were 200 μs in duration or 1, 2, or 3 mA with phases that were 500 μs in duration.

When stimulation was conducted at 50 Hz with an anodal phase duration of 500 μs the right neck contracted at a current strength of 3 mA in electrodes 1-4 and 7-8, but at a strength of 2 mA for electrodes 5-6. When the lead phase was cathodal, then the right neck contraction was evident at 2 mA in all electrodes. Similarly, higher currents were at times required with an anodal lead phase at 50 Hz when the phase durations were 200 μs. Electrodes 3-4 and 7-8 induced contractions at 6 mA, whereas electrodes 1-2 and 5-6 induced contractions at 3 mA. In contrast, when a cathodal lead phase was used, contractions were induced at 3 mA across all electrodes.

Results were similar when the frequency was 20 Hz. Phase durations of 500 μs induced right neck contractions at 2 mA when the lead phase was cathodal, but when it was anodal electrodes 1, 3, and 5-7 needed a current of 3 mA to induce the contraction. Phase durations of

200 μs induced right neck contractions at 3 mA when the lead phase was cathodal, but when it was anodal electrodes 1, 3-5 and 7-8 needed a current of 6 mA to induce the contraction.

1.1.5 B63

1.1.5.1 Experiment #1

B63 was first stimulated whilst standing in its metabolism cage; this cage was positioned in the sheep pen so that it was in the company of other sheep. No distress was evident for the sheep during the stimulation studies and it appeared to be in good condition.

The stimulation configuration available was restricted to monopolar stimulation, using any one of the seven electrodes in combination with the rear ground as the return. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse, either as an anodal or cathodal current, with a 200 μs or 500 μs phase duration. Currents used were either 3 or 6 mA with phases that were 200 μs in duration or 1, 2, or 3 mA with phases that were 500 μs in duration.

Stimulations at 50 Hz with a 500 μs phase width were not associated with any activity. Then 20 Hz and a 500 μs phase with was used, but again without a response. However, when a 200 μs phase width was used at a current of 6 mA, but no at 3 mA, and at either 20 Hz or 50 Hz, electrode 7 alone was associated with fasciculations in the left supra lip region. This occurred when the electrode acted as the cathode, but not when it was an anode, and the threshold appeared to be 4.5 mA.

The head return was not used, nor were bipolar configurations trialled.

1.1.5.2 Experiment #2

In order to replicate the results of the first experiment, the sheep was placed in a sling and tethering equipment, comprised of the head collar attached. The sheep tolerated the transfer well. Acupuncture needles were positioned subcutaneously.

Stimulations were performed through electrode 7 alone, in a monopolar configuration using either the head or the rear return. The frequencies used were both 20 Hz and 50 Hz with 200 μs phase widths and a current of 6 mA for 3 seconds. These led to fasciculations of the left supralip region as before. In addition, a 1 Hz stimulation was trialled for a run of 10 seconds, but with no effect. The electromyography acupuncture needles detected stimulation artefact alone and no objective evidence that contractions had taken place.

1.1.5.3 Experiment #3

To gather objective evidence of the left supra-lip region fasciculations, the sheep was again placed in a sling with a head collar attached and accelerometers secured in place on the left and right supra-lip regions.

Stimulations were performed through electrode 7 alone, in a monopolar configuration using the rear return. The frequency used was 20 Hz with a 200 μs phase width and a current of 6 mA for 3 seconds. This led to fasciculations of the left supra-lip region as before. The accelerometers documented the fasciculations.

1.1.5.4 Experiment #4

All seven electrodes had been studied on one occasion in B63, and using the rear return alone. As such the sheep was again placed in a sling with tethering equipment attached and accelerometers to the left and right facial regions.

Stimulations were performed through all seven electrodes, in a monopolar configuration using the head return. The frequency used was 20 Hz with a 200 μs phase width and a current of 6 mA for 3 seconds. Using this configuration, the first electrode was associated with a contraction in the left neck region. Then, the second to fifth electrodes were each associated with left facial contractions, which had not occurred previously when the rear return was used. The sixth electrode was not associated with any contraction at all. The seventh electrode still induced a left supra-lip fasciculation. While it had been planned to immediately compare these findings with the rear return, the sheep became restless and was removed from the sling and returned to its cage.

1.1.5.5 Experiment #5

To compare the effect of return used in a monopolar configuration as well as responses to bipolar configurations, the sheep was again placed in a sling, but with no accelerometer monitoring.

Stimulations were performed using a frequency of 20 Hz conducting a 200 μs either anodal or cathodal monophasic pulse for 3 seconds. When the head return was used in a monopolar configuration, the first electrode had no response when acting as the anode, but when it was the cathode the left neck contracted. The second electrode was associated with left neck contractions when acting as either the anode or cathode. The third and fourth electrodes had no response when acting as the anode, but induced left supra- and infra-lip fasciculations when acting as the cathode. The fifth and sixth electrodes had no effect when acting as the anode, but induced left neck contractions when acting as the cathode. The seventh electrode induced left neck contractions when acting as the anode or cathode.

When the rear return was used, the results were similar. The first electrode caused left neck contractions when acting as the cathode, but no anode. The second electrode had no effect at all. The third and fourth electrodes had no effect as the anode, but caused left supra- and infra-lip fasciculations as a cathode. The fifth electrode had no effect at all. The sixth electrode caused left neck contractions when acting as either the anode or cathode. The seventh electrode had no effect at all.

Bipolar configurations, where electrode 1 returned via 2, then 2 via 3 and so on, were not associated with any contractions except for when electrode 1 and 2 were used and 2 and 3; these configurations were associated with left neck contractions. In addition, a doublet bipolar setup was trialled where the current was set to 10 mA. The only response noted was when a doublet of electrodes 1 and 2 were returned via a doublet of 3 and 4, which consisted of a left neck contraction.

1.1.6 B65

1.1.6.1 Experiment #1

B65 was stimulated whilst in a sling, but without tethering equipment. There were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place four days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or the rear return used. The parameters trialled included a frequency of 20 Hz stimulating for 3 seconds with a monophasic pulse, with the electrode acting as either an anode or cathode, and with a 200 μs phase duration. Current strength was set to 6 mA.

The bipolar configurations were not associated with any response. When the rear return was used there were also no responses. Then, when the head return was used, for each of the twelve electrodes a response about the left ear was noted where it would retract and fasciculate.

1.1.6.2 Experiment #2

To explore additional stimulation parameters B65 was again stimulated whilst in a sling, without tethering equipment. There were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place thirteen days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with the rear return used. The parameters trialled included a frequency of 20 Hz stimulating for 3 seconds with a monophasic pulse, with the electrode acting as either an anode or cathode, and with a 500 μs phase duration. Current strength was set to 3 mA.

The bipolar configurations were not associated with any response. When the rear return was used there were also no responses.

Then, a double consisting of electrodes one and two was used in a monopolar fashion with either the head or rear return used and whilst stimulating with a current of 10 mA. When this doublet acted as a cathode, both forelegs contracted when the rear return was used, but not when the head return was used. No effect was noted when this doublet acted as the anode with either return.

Doublet bipolar configurations, consisting of electrodes 1 and 2 as a doublet connected to either a doublet of 6 and 7 or a doublet of 11 and 12, were not associated with any responses.

1.1.6.3 Experiment #3

To trial additional stimulation parameters B65 was again stimulated whilst in a sling, without tethering equipment. Several days before these studies, the sheep's head had been placed in a stereotactic frame for another experiment, and consequently suffered a left facial palsy. In spite of this, there were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place 25 days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or rear return used. The parameters trialled included a frequency of 50 Hz stimulating for 3 seconds with a monophasic pulse, with the electrode acting as either an anode or cathode, and with a 200 μs or 500 μs phase duration. Current strength was set to 6 mA for 200 μs phases and 3 mA for 500 μs phases.

Bipolar configurations were not associated with any responses. Then, monopolar stimulation was used and no responses were noted when either the head or rear return was used.

1.1.6.4 Experiment #4

To explore 1 Hz pulse trains B65 was again stimulated whilst in a sling; no tethering equipment was used.

Stimulation took place 28 days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or rear return used. The parameters trialled included a train of 5 monophasic pulses, each pulse of 200 μs duration and these at a frequency of 2,500 Hz. This train of pulses was repeated at a frequency of 1 Hz stimulating for 10 seconds; the electrode acted as either an anode or cathode and the current strength was set to 6 mA.

While there were no responses when bipolar configurations were used, or the head return, when the rear return was used, electrodes 4, 6, 10, 11, and 12 were associated with contraction of the right and left neck when acting as the cathode, but not as the anode.

1.1.7 B66

1.1.7.1 Experiment #1

B66 was stimulated whilst in a sling, but without tethering equipment. There were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place four days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or the rear return used. The parameters trialled included a frequency of 20 Hz stimulating for 3 seconds with a monophasic pulse, with the electrode acting as either an anode or cathode, and with a 200 μs phase duration. Current strength was set to 6 mA.

None of the configurations trialled were associated with any activity in B66.

1.1.7.2 Experiment #2

To explore additional stimulation parameters B66 was again stimulated whilst in a sling, without tethering equipment. There were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place thirteen days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or rear return used. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse. The electrode acted as either an anode or cathode, and a 200 μs or 500 μs phase duration was used. Current strength was set to 6 mA for 200 μs phases and 3 mA for 500 μs phases.

None of the bipolar configurations were associated with muscular activity. A pulse train parameter, as has been described, was trialled with electrodes 1 and 8 in a bipolar configuration alone, but there was no response. In addition to single electrode bipolar configurations, doublet bipolar stimulation was trialled with currents of 10 mA at 20 Hz and 200 μs phase durations. A doublet of electrodes 1 and 2 connected to either a doublet of electrodes 4 and 5 or 7 and 8 was not associated with any response.

Monopolar configurations using the rear return and any one of the eight electrodes from the device was associated with fasciculations around the return when the electrodes of the device acted as the anode. This was also the case when a doublet of electrodes 1 and 2 or 7 and 8 were used with the rear return. When a pulse train stimulation parameter was trialled with electrode 1 connected to the rear return, there was no response.

Monopolar configurations to the head return were not associated with any activity.

1.1.7.3 Experiment #3

To explore 1 Hz pulse trains B66 was again stimulated whilst in a sling; no tethering equipment was used.

Stimulation took place 22 days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or rear return used. The parameters trialled included a train of 5 monophasic pulses, each pulse of 200 μs duration and these at a frequency of 2,500 Hz. This train of pulses was repeated at a frequency of 1 Hz stimulating for 10 seconds; the electrode acted as either an anode or cathode and the current strength was set to 6 mA.

There were no responses when a bipolar configuration was used. However, when a monopolar configuration was used with the rear return, each of the eight electrodes induced a left neck contraction when it acted as a cathode, and when it acted as an anode, there were fasciculations about the return site. In contrast, there were no responses to monopolar stimulations where a head return was used.

1.1.8 B67

1.1.8.1 Experiment #1

B67 was stimulated whilst in a sling and without tethering equipment. There were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place four days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or the rear return used. The parameters trialled included a frequency of 20 Hz stimulating for 3 seconds with a monophasic pulse, with the electrode acting as either an anode or cathode, and with a 200 μs phase duration. Current strength was set to 6 mA.

None of bipolar or monopolar to head return configurations were associated with a response. When the rear return was used, and any of the eight electrodes was acting as the anode, there were fasciculations about the return. Then, when they acted as the cathode, there were bilateral neck contractions.

1.1.8.2 Experiment #2

To explore additional stimulation parameters B67 was again stimulated whilst in a sling, without tethering equipment. There were no signs of distress and it appeared that the animal tolerated the stimulations studies well.

Stimulation took place thirteen days after the stent-electrode array had been implanted. The stimulation configurations included bipolar and monopolar orientations with either the head or rear return used. The parameters trialled included a frequency of either 20 Hz or 50 Hz stimulating for 3 seconds with a monophasic pulse. The electrode acted as either an anode or cathode, and a 200 μs or 500 μs phase duration was used. Current strength was set to 6 mA for 200 μs phases and 3 mA for 500 μs phases. In addition, a train of 5 monophasic pulses, each pulse of 200 μs duration and these at a frequency of 2,500 Hz was used. This train of pulses was repeated at a frequency of 1 Hz stimulating for 10 seconds; the electrode acted as either an anode or cathode and the current strength was set to 6 mA.

In the bipolar configuration, when electrode one was paired to any of the other electrodes, it caused bilateral limb contractions at 50 Hz with a phase duration of 200 μs at 6 mA; a more pronounced response was noted when it acted as the cathode. This response was not noted when electrode 2 was paired to the other electrodes except for 1 and 3, when it was. All other bipolar configurations weren't associated with a response. With the same frequency, but using a phase of 500 μs at 3 mA, bipolar configurations that induced this response were restricted to electrode 1 paired to either 2 or 3 alone, but not when it acted as the anode. There were no responses in the bipolar configuration when the frequency was changed to 20 Hz.

When a monopolar configuration was used with a frequency of 20 Hz and phases of 200 μs, electrodes 1-3 were associated with left neck contractions and then fasciculations about the rear return when they acted as the cathode and anode respectively. However, when the phase duration was 500 μs with either 50 Hz or 20 Hz the responses were not elicited. A frequency of 50 Hz, but with a phase duration of 200 μs, was again associated with the same responses. The head return was not associated with any responses with all parameters trailed.

Bipolar configurations, in association with the pulse train stimulation parameters were intermittently associated with left and eye lid twitches, more pronounced on the right than on the left.

Unanswered Questions

1) Were the contractions induced consequences of motor cortical stimulation or current spread?
2) What is the current range likely to effectively stimulate the Ovine motor cortex?
3) Are larger electrodes required in order to facilitate greater current strengths; could a stent carry larger electrodes?
4) Does stimulation of the sinuses, bound by the dura, cause pain?
5) At what time point do the cables connecting the stent-electrode arrays to their external plugs break?
6) Does stimulation of the dura lead to contraction of the sinus?

What these research outcomes mean

In essence, this body of work lays the foundations for future stimulation studies. At this stage the positive steps taken include the reliable fabrication of device and method to deploy it, and now a stimulation paradigm with equipment to facilitate the experiments. However, future work will need to focus on bipolar stimulation configurations (to avoid current spread) and employ higher currents. In addition, the spread of current that the device can carry needs to be documented *in vivo*.