Electroencephalography as a method to distinguish between pain and anaesthetic intervention in conscious lambs undergoing castration

C. Harris [A,C], V. L. Mohler [B], P. White [B] and S. Lomax [A]

[A]School of Life and Environmental Sciences, Faculty of Science, The University of Sydney, Australia.
[B]Sydney School of Veterinary Science, Faculty of Science, The University of Sydney, Australia
[C]Email: char2922@uni.sydney.edu.au

Australian sheep routinely undergo painful surgical husbandry procedures without anaesthesia or analgesia. This includes castration which has been demonstrated to illicit pain related behaviour in lambs (Lester et al. 1996). A successful measure of pain in livestock under a general anaesthetic is electroencephalography (EEG) using a minimal anaesthesia method (MAM) (Murrell and Johnson 2006). However, this method has many practical limitations including constraints in field settings, cost and time restrictions. Conscious EEG recording has the potential to overcome these limitations. This study compares the EEG output of the MAM with that of conscious lambs undergoing castration with and without local anaesthesia. Sixteen merino crossbred ram lambs were randomly allocated to 1 of 4 treatment groups: namely, 1) conscious EEG and surgical castration with no anaesthetic intervention (CON; n=4); 2) conscious EEG and surgical castration with pre-operative applied intra-testicular lignocaine injection (CON+LIG; n=4); 3) surgical castration under minimal anaesthesia (MAM; n=4); and 4) surgical castration with pre-operative lignocaine injection, using 2mL of a 20mg/mL lignocaine hydrochloride solution, under minimal anaesthesia (MAM+LIG; n=4). The EEG output was analysed using MATLAB (MathWorks, Inc. version R2017b) to calculate total power (Ptot), median frequency (F50) and spectral edge (F95). A restricted maximum likelihood test (REML) was also conducted using R (Version 1.1.447 – © 2009-2018 RStudio, Inc.). P values < 0.05 were considered statistically significant. Significant changes were observed for all treatments from Pre- to post- surgical values with the exception of F50 MAM+LIG (Table 1). Furthermore, CON and CON+LIG treatment groups were distinguishable using F50 (p=0.02) and F95 (p=0.04) measurements post castration (Table 2). The use of EEG was successful in differentiating conscious, castrated lambs treated with pain relief from those without any treatment, by examining the F50 and F95 parameters, suggest possible suitability of conscious EEG for pain measurement.

	Time	CON (1)	CON+LIG (2)	MAM (3)	MAM+LIG (4)
F50	Pre	2.45(11.61)	2.63(13.81)	2.21(9.08)	2.23(9.33)
	post	2.41(11.11)	2.68(14.60)	2.28(9.74)	2.24(9.36)
	p value	<0.01	<0.01	<0.01	0.7493
F95	Pre	3.13(22.87)	3.18(23.96)	2.96(19.29)	2.98(19.73)
	post	3.08(21.85)	3.17(23.71)	3.00(20.05)	3.00(20.04)
	p value	<0.01	<0.01	<0.01	<0.05
Ptot	Pre	$-9.44(7.97\times10^{-5})$	$-9.53(7.25\times10^{-5})$	$-8.97(1.27\times10^{-4})$	$-9.24(9.71\times10^{-5})$
	post	$-9.25(9.63\times10^{-5})$	$-9.63(6.59\times10^{-5})$	$-8.88(1.39\times10^{-4})$	$-9.10(1.12\times10^{-4})$
	p value	<0.01	<0.01	<0.01	<0.01

Table 1: Predicted means, with back-transformed values in parenthesis, for each treatment group before (Pre) and after (Post) castration

		Treatment comparisons					
		1-2	1-3	1-4	2-3	2-4	3-4
F50	Pre	0.12	0.04	0.06	<0.01	<0.01	0.80
	Post	0.02	0.22	0.12	<0.01	<0.01	0.70
P95	Pre	0.22	<0.01	<0.01	<0.01	<0.01	0.55
	Post	0.04	0.03	0.03	<0.01	<0.01	0.99
Ptot	Pre	0.82	0.26	0.63	0.18	0.48	0.51
	Post	0.36	0.37	0.71	0.08	0.20	0.60

Table 2: P-values for pairwise comparisons of predicted means between treatment groups, within a time period, pre or post castration. Treatment groups are denoted as follows: Con (1), Con+Lig (2). MAM(3), MAM+LIG (4)

References

Lester SJ, Mellor DJ, Holmes RJ, Ward RN and Stafford KJ (1996) *Behavioural and cortisol responses of lambs to castration and tailing using different methods: New Zealand Veterinary Journal,* vol. 44, no. 2, pp. 45-54.
Murrell JC and Johnson CB (2006) *Neurophysiological techniques to assess pain in animals: Journal of Veterinary Pharmacology and Therapeutics.* **29**(5) 325-335.

The authors acknowledge and thank Australian Wool Innovation for funding this work.