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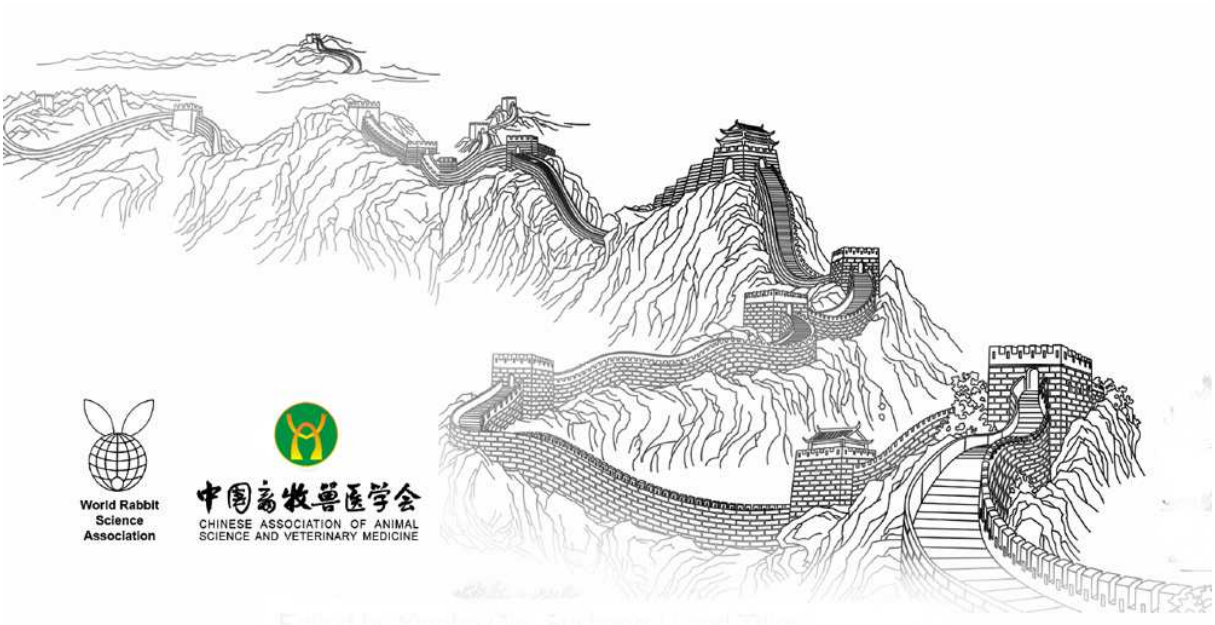

DEFINITION OF INDICATORS TO EVALUATE CONSCIOUSNESS OF RABBITS AT THE TIME OF SLAUGHTER AND OPTIMISATION OF PARAMETERS FOR ANIMAL PROTECTION

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ABSTRACT

Stunning is an important stage of the slaughter process, which directly influences the following steps and also the final quality of meat and products. Regulation 1099/2009 on animal protection at the time of killing raises a positive list of stunning methods. In rabbits, the most common method is head-only electrical stunning. However, studies have shown that electrical parameters could be optimised to guarantee unconsciousness of animals up to death. The aim of our study was first to propose practical and reliable indicators of consciousness for industrial conditions, and also to determine, using indicators of consciousness under experimental conditions, optimal parameters of narcosis (frequency, voltage, intensity and time of application) to guarantee unconsciousness of rabbits from stunning to death. The tracking of individual rabbit batches in several slaughterhouses enabled us to propose a set of practical indicators to estimate consciousness of animals. Thus, just after stunning, tests of awakening by measuring the time before the corneal reflex reappears and possibly posture seem to be relevant. Observing open eyes just after electro-narcosis, although less precise, also seems to be usable and allows the observation of a larger number of animals. In the light of the results of this study it is recommended that to validate the unconsciousness of animals at bleeding, the corneal reflex test and observing for eye blinking, should be carried out on the chain on a minimum of 1 to 3 % of rabbits per batch. To guarantee unconsciousness of rabbits during the slaughter process the experimental phase determined that, with the tested parameters, bleeding should take place within 30 seconds of electro-narcosis. The best result for the quality of unconsciousness was obtained with electrical stunning for one second whatever the electrical parameters.

Key words: Rabbit; Welfare; Electro-narcosis; Stunning; Slaughter

INTRODUCTION

Animal stunning at slaughterhouse is an essential stage before bleeding and allows the animal to be maintained in a state of unconsciousness until death. To supervise this practice, the European Union established regulation 1099/2009 on the protection of animals at the time of killing (EU, 2009), applicable since January 2013. Unlike for other species, this regulation for rabbits does not indicate minimum electric parameters or duration of current application.

In rabbits, electrical stunning is the most common method used. However, studies have shown that electric parameters could be optimised to guarantee unconsciousness of animals up to death. Studies on the impact on animal welfare of electro-narcosis parameters are relatively few in rabbits. Although concerning few animals, the reappearance of consciousness before death indicates that, in certain cases, stunning duration is insufficient. Electrical parameters of electro-narcosis seem to affect the efficiency and duration of anaesthesia. Maria *et al*. (2001) showed that applying a low voltage (19V) could lead to a lack of anaesthesia for some animals. In this study, the current was applied for three seconds and frequency was varied between 161 Hz and 1,667 Hz resulting in no noticeable impact on the quality of anaesthesia. Anil *et al*., (1998) also showed that the voltage applied and the duration of application could have an effect on the quality of the anaesthesia and the time to regain consciousness. Higher voltages and time of application resulted in more efficiency stunning. Anil *et al*. (2000) showed that with a frequency

of 50 Hz and an application of 1 second the amount of current also affected the brain's response to electro-narcosis and they concluded that a minimum current of 140 mA should be used. They also showed that not all of the rabbits presented epileptic reactions seen by tonic-clonic phases characterised by rigidity followed by motor activity normally characterised by spontaneous leg jerks. Thus, in spite of contrasting results in the literature, it seems that electrical parameters of narcosis influence its quality and the time between when the electrical shock is applied and the animal regaining consciousness.

The aim of our study was first to propose practical and reliable indicators of consciousness for industrial conditions, and also to determine, using indicators of consciousness under experimental conditions, optimal parameters of narcosis (frequency, voltage, intensity and time of application) to guarantee unconsciousness of rabbits from stunning to death.

MATERIALS AND METHODS

Survey of slaughterhouses to determine indicators of consciousness

Observations were conducted in three slaughterhouses in France, on 16 batches of rabbits (900 animals per hour), whatever their genotypes. Observations were made at electro-narcosis and bleeding locations and mean application time and mean intensity of current were calculated for 10 animals. Indicators that can be observed at slaughter were counted twice during four minutes at narcosis, neck cutting and during bleeding. The awakening tests were performed just after electro-narcosis. Indicators observed were open eyes, blinking eyes, neck tension, vocalizations, backpedalling of the front legs and gasping. Results of the measures were converted into percentages. Finally, awakening tests, as described below, were practised on 10 animals chosen at random.

Determination of optimal electric parameters

Six experimental modalities of electro-narcosis were tested on rabbits. Two voltages and three durations of application were tested. At 50 Hz, rabbits were subjected to a voltage of 200 or 300 V, for 0.5, 1 or 2 seconds. Two different procedures were used to test rabbit unconsciousness:

- An awakening test, on 4 rabbits per condition (24 rabbits in all), were stunned and then placed individually in boxes to measure their awakening time. The time before reappearance of the corneal reflex and then of the posture were recorded during the first two minutes after electro-narcosis.
- A test was conducted on the slaughter chain, with 36 rabbits for each of the 6 conditions (216 rabbits in all) stunned then bled. The unconsciousness of rabbits was estimated using the corneal reflex test, spontaneous blinking eyes, gasping and vocalizations observed between electro-narcosis and bleeding and then 15 seconds after bleeding.

Statistical Analysis

Normality of data was verified with the PROC UNIVARIATE of SAS and using a graphic approach. In determining indicators, data did not follow a normal distribution and Spearman rank correlations were used as a non-parametrical alternative to compare the consciousness indicators. Tests of Mann Whitney and Kruskal Wallis (proc NPar1Way) were used to highlight the main effects. For the evaluation of the electrical parameters, normal data were processed using an analysis of variances (Anova), followed in the case of significant differences by Fisher tests. For the non-normal data, non-parametrical tests of Kruskal Wallis were performed, when significant differences were revealed, pairwise Mann-Whitney tests were conducted with Bonferroni correction.

RESULTS AND DISCUSSION

Field survey to determine consciousness indicators

Descriptive statistics:

The survey in the French slaughterhouses highlights that the frequencies used were 50 Hz or 200 Hz with voltages of 300 or 110 V respectively. The average body weight of rabbits was between 2.33 and 2.55 kg. Intensity received by each animal was of 360 mA on average and it varied from 71 to 870 mA.

Correlation between indicators:

Positive correlations were observed between corneal reflex and resumption of posture. In addition, as the time for the corneal reflex to reappear and posture to be resumed increased, the number of animals with open eyes after electro-narcosis decreased (rho = -0.83 and -0.87 respectively) and the animals which backpedalled during bleeding rose (rho = 0.57 with corneal reflex). A negative correlation was observed between the indicators of "open eyes after electro-narcosis" and "tension of the neck at electro-narcosis" (rho = -0.82). Most indicators used at electro-narcosis and during bleeding were not correlated except for tension of the neck after electro-narcosis and backpedalling during bleeding (rho = 0.75). Positive correlations were found between various indicators of consciousness observed during bleeding. The percentage of animals which backpedalled was positively correlated with the percentage of animals which blinked during bleeding (rho = 0.70), the latter being positively correlated with the percentage of gasping (rho = 0.69). Vocalizations at and during bleeding were also positively correlated (rho = 0.81).

Discussion and definition of a set of indicators: V

ery few vocalizations were heard in rabbit slaughterhouses making it difficult to use as a relevant indicator. However, when vocalizations were heard after hanging, it revealed a problem of narcosis. The positive relation between corneal reflex and resumption of posture is logical because of their positive correlation with duration of unconsciousness. These two tests thus appear appropriate. On the other hand, there was a positive relationship between corneal awakening test and the percentage of animals which backpedalled which is surprising because these indicators should not measure the same thing. This could indicate that animals which backpedalled were well stunned. However, there is a positive relationship between backpedalling and blinking eyes during bleeding which would indicate that animals which express this reaction are conscious. This could be due to the difficulty discriminating between backpedalling reflexes linked to the clonic phase of epilepsy and the voluntary backpedalling possibly indicating an attempt to escape and thus suffering. Furthermore, the positive relationship between gasping and blinking eyes, signs of an animal's consciousness, is in line with the conclusions of Rota Nodaris et al. (2009) on the negative significance of gasping. Parameters of consciousness observed at electro-narcosis were in no way correlated with those observed at bleeding. For Anil and al. (1998, 2000) and Maria and al. (2001), the best indicator of unconsciousness of animals is the respiratory arrest which follows the application of the current. A well stunned rabbit should stop breathing for 22 seconds. However, in practice this indicator is difficult to observe whether during an awakening test or on the slaughter chain. Maria and al. (2001) and Rota Nodaris and al. (2009) cite both of these difficulties. For these authors, corneal reflex is a relevant indicator which is simpler to operate in the field, which is confirmed by our work.

Determining parameters of optimization of narcosis

Awakening test: effect of electrical parameters on the state of unconsciousness of rabbits:

Results were too variable to highlight an effect of the voltage or duration of application on the corneal reflex and posture tests (Table 1). On the other hand, there was a tendency that, whatever the parameters of electro-narcosis, 100 % of rabbits had a tonic phase after stunning, and that corneal reflex always returned after a minimum of 28 seconds after electro-narcosis which did not lead to the death of the animal (96 % of rabbits were conscious 2 min after electro-narcosis).

Effect of electrical parameters on the state of unconsciousness of rabbits between electro-narcosis and bleeding and after bleeding:

For our study, between electro-narcosis and bleeding, voltage and duration of application had no effect on spontaneous blinking, corneal reflex and gasping. Fifteen seconds after bleeding, voltage and duration of application had no effect on corneal reflex and gasping. On the other hand, spontaneous blinking after bleeding was more frequent for a voltage of 300 V and for an application time of 0.5 second (Table 2).

Table 1: Effects of the voltage and application duration on the corneal reflex and posture tests

Treatment	Corneal reflex (s)	Posture test (s)	Number of rabbits
200 V	37	76	12
300 V	45	90	
SEM	6	6	
P	**NS**	**NS**	
0,5 s	48	83	8
1 s	38	80	
2 s	36	86	
SEM	7	7	
P	**NS**	**NS**	

Table 2: Effect of voltage and duration of narcosis on spontaneous blinking, corneal reflex and gasping after heavy loss of blood

Treatment	Blinking eye - Bleeding	Corneal reflex - Bleeding	Gasp - Bleeding
200 V	4/119	31/119	6/119
300 V	16/119	30/119	2/119
P	**< 0,01**	**NS**	**NS**
0,5 s	15/80	24/80	2/80
1 s	3/78	21/78	4/78
2 s	2/80	16/80	2/80
P	**< 0,001**	**NS**	**NS**

CONCLUSIONS

Our study has enabled us to recommend a set of practical indicators that can be used in slaughterhouses to estimate the consciousness of rabbits. Just after stunning, an awakening test timing the return of corneal reflex and then posture appear to be relevant. In the case of retaining only one of these tests, the corneal reflex is more appropriate. Although less precise, observing open eyes just after stunning, is also possible and allows a larger number of animals to be observed. In the light of the results obtained in this study, to validate unconsciousness of animals during bleeding, the corneal reflex test should be conducted on the slaughter chain on a minimum of 1 to 3 % of the rabbits per batch. Observation of blinking eyes on the same number of rabbits (1 to 3 %) during bleeding could also be a satisfactory criterion.

In terms of optimal parameters, in our experimental conditions, the first results with the parameters tested show that bleeding should take place within 30 seconds of electro-narcosis to guarantee unconsciousness of rabbits during their slaughter. An application time of one second seems to give the best results for the quality of unconsciousness.

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