

Paper

Forensic cases of bruises in pigs

K. Barington, H. E. Jensen

Bruises in pigs inflicted by blunt trauma are a significant animal welfare problem, and affected skin and underlying muscle are regularly submitted for forensic investigation. Central to the evaluation is an assessment of the age of the bruises. This paper presents cases of bruises in pigs sent for forensic investigation that were collected retrospectively. Data comprised photographs of the gross lesions, slides for histology, and written reports. The time from collecting the animals at the farms and delivery to the slaughterhouse was recorded together with the time of slaughter. Since 2005 there has been an increase in cases, with a peak in 2008 and 2009 of 40 cases for each year. At gross examination, the pattern of bruises often reflected the type of object which caused them. Histologically, haemorrhage and cellular infiltrations were frequently present. Currently, the age of bruises may be estimated to be more or less than four hours based on a porcine bruise model. In bruises more than four hours old, estimations of two-hour intervals are used based on studies of wound healing. The time from collecting the pigs at the farms until slaughter was between one and four hours in 44.1 per cent of cases, during which time the pigs had been handled by several people. In addition, in 22.0 per cent of cases of bruising an inflammatory response was absent, making it impossible to estimate the age of the bruise.

Introduction

Assessment of the age of bruises is a central issue in human forensic pathology (Vanezis 2001). This has also become increasingly important in cattle and pigs (McNally and Warriss 1996, Dalla Costa and others 2007, Huertas and others 2010, Hoffman and Luehl 2012). A bruise is defined as a visible extravasation of erythrocytes in the subcutis and surrounding tissue due to trauma with a blunt instrument. The impact from a blunt object leaves the skin surface intact while the walls of veins, venules and small arteries are torn, leaking blood into the surrounding tissue (Hamdy and others 1957, Langlois and Gresham 1991, Saukko and Knight 2004). Information on the role of bleeding from capillaries is contradictory. For example, Saukko and Knight (2004) state that capillary bleeding is not visible on gross examination, while Langlois and Gresham (1991) define a bruise as being due to bleeding from capillaries and small veins.

This paper presents cases of bruises in Danish slaughter pigs and sows sent for forensic investigation. Typical gross and histological findings in bruises of pigs and the increase in the number of cases during the last eight years are reported. In addition, the challenges associated with estimating the age of the bruises are discussed.

Materials and methods

Cases of bruises in pigs sent for forensic investigation at the University of Copenhagen were collected retrospectively. Cases of blunt trauma in pigs received from January 2005 to May 2013 were included. All cases were identified by veterinarians at the postmortem inspection at 14 slaughterhouses in Denmark. In all cases, the data comprised

photos of the gross lesions, slides for histology, and written reports. The time from collection of the animals at the farm to delivery at the slaughterhouses was recorded together with the time of slaughter.

In each case, skin and muscle tissue was sampled for histology and stained with haematoxylin and eosin. In most cases (more than 90 per cent), the carcasses were scalded, singed and scraped before tissue sampling. To avoid decomposition, bruised skin and muscle were frozen before submission for forensic investigation. In some cases though, bruised tissue was sampled before freezing of the pigs and preserved in 10 per cent formalin solution.

Results

Since 2005 when a single case was examined, the number of cases has increased and peaked in 2008 and 2009, with 40 cases being recorded in each year (Fig 1). Thereafter, the annual number of cases has decreased and in 2012 a total of 16 cases were examined (Fig 1). During the first five months of 2013, only three cases of bruises in pigs

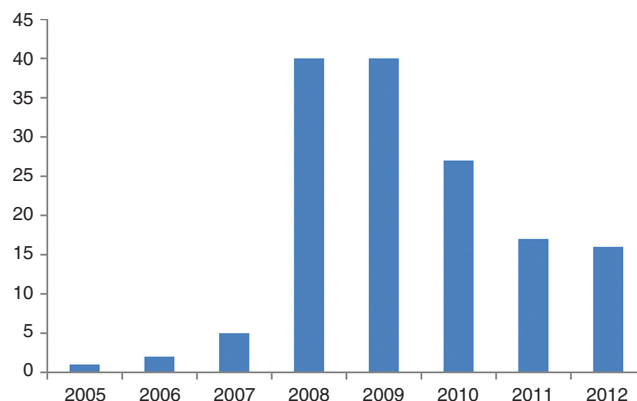


FIG 1: The annual number of forensic cases concerning bruises in pigs. The horizontal axis indicates the year, and the vertical axis indicates the number of cases

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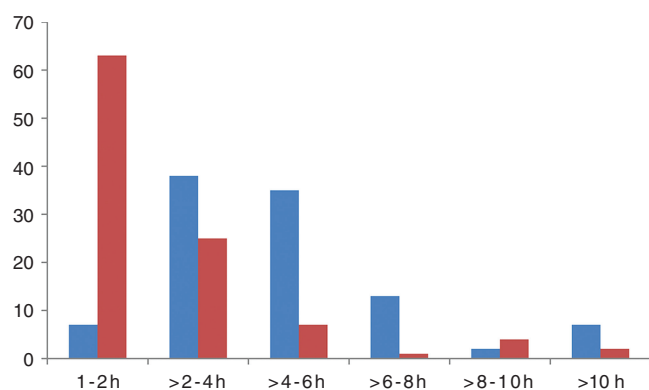


FIG 2: Periods of time that elapse between collection of animals from the farm and their slaughter (blue bars), and between delivery at the slaughterhouse and their slaughter (red bars). The horizontal axis indicates the timespan, and the vertical axis indicates the number of cases

due to blunt trauma have been received. In all cases the bruises were found on white pigs (Landrace crossbred), which is the dominant type of pig slaughtered in Denmark.

In 75 of 148 cases, details on the number of pigs involved were available. In summary, 309 pigs in total were bruised in these cases. In 93.33 per cent of the cases 1–10 pigs were involved, while in the remaining cases up to 42 pigs were involved. In 87 of 148 cases, information regarding the farms was available. In these cases a total of 85 farms were involved, three farms were involved in more than one case, and in two cases the affected pigs were delivered from two different farms.

In 102 of 148 cases, information regarding the time of collection of the animals at the farm, the time of delivery to the slaughterhouse, and the time of slaughter was available (Fig 2). The minimum and maximum time periods from when the pigs were collected until they were slaughtered were one hour and 15 minutes, and 23 hours and 25 minutes, respectively. The timespan between collection of the animals and slaughter was four hours or less in 44.1 per cent of cases, and between four and eight hours in 47.1 per cent. In the remaining cases (8.8 per cent), the timespan between collection and slaughter was more than eight hours. The minimum timespan from delivery of the pigs to the slaughterhouse and their slaughter was five minutes, and the maximum timespan was 18 hours and 55 minutes. In the majority of cases (86.3 per cent), the timespan between delivery and slaughter was four hours or less. In the remaining cases (13.7 per cent), a timespan of more than four hours between delivery and slaughter was recorded.

In all cases, the bruises were localised on the back of the pigs. At gross examination, bruises typically had a tram-line appearance indicating that the trauma was inflicted by a stick (Fig 3). Other bruises were presented as confluent accumulation of blood in the skin and

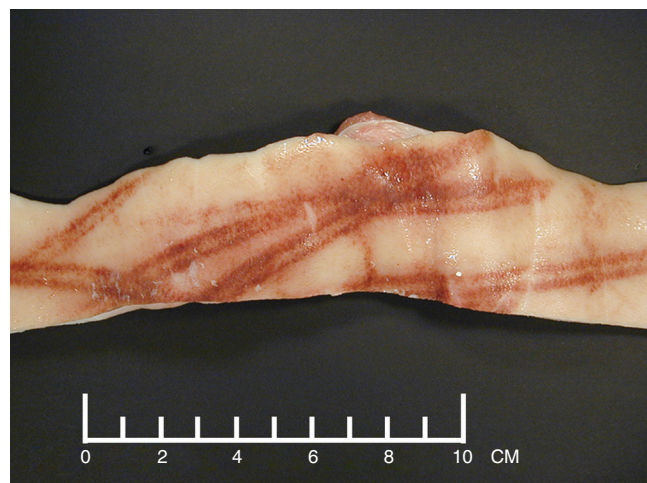


FIG 3: Skin of a pig with bruises with a tram-line appearance

some had a pattern that reflected the tools or chains by which they were inflicted (Figs 4 and 5). By gentle palpation, bruises were found to be slightly raised compared to the surrounding skin. Following cross-sectioning of the skin, the bruises were sometimes found to extend into the subcutaneous tissue and in several cases further into the underlying muscle tissue (Fig 6).

The processing of the carcass had removed the epidermis and resulted in the transformation of the dermis to a homogeneous mass at microscopic examination. In addition, freezing the tissue made the erythrocytes burst, thereby making them difficult to recognise during microscopic evaluation (Fig 7). In cases where bruised tissue was sampled before freezing the erythrocytes had a normal appearance (Fig 8). In the subcutaneous tissue, extravasated erythrocytes were observed between the adipocytes but especially along the fibrous septa that are longitudinally arranged in the deep subcutis of pigs. The extent of changes varied from clusters of a few erythrocytes to larger areas of haemorrhage (Figs 7 and 8). In relation to the erythrocytes, infiltrating neutrophils and macrophages were often present to a variable extent (Figs 7 and 8). Changes in the muscle tissue were sometimes absent even though the most significant and advanced changes were typically seen in the muscle tissue. Lesions were present in the form of single necrotic muscle fibres surrounded by normal myofibres, or as clusters of necrotic myofibres (Figs 9 and 10). Necrotic myofibres were often swollen and fragmented (Fig 10). The necrotic myofibres had pale, homogenous cytoplasm compared to viable myofibres. Cut longitudinally, necrotic fibres showed a loss of striation (Fig 11). Necrotic muscle fibres were accompanied by infiltration of neutrophils and macrophages (Figs 11 and 12). The inflammatory cells were present to

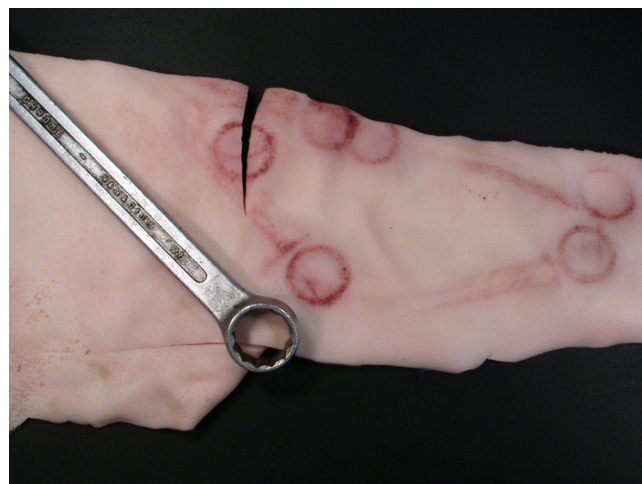


FIG 4: Bruises on the skin of a pig caused by impact from the tool (spanner) shown in the picture



FIG 5: Bruises in pig skin caused by impact from the chain shown in the picture

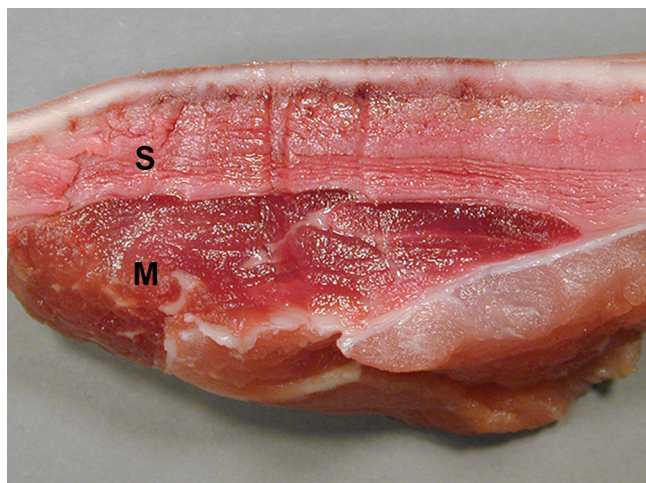


FIG 6: Cross-section of skin (S) and muscle (M) from a pig subjected to blunt trauma. Haemorrhage is seen in all layers of the skin and in the underlying muscle

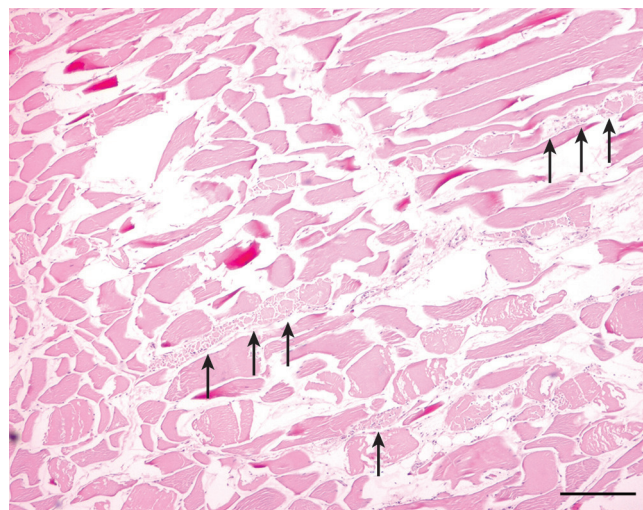


FIG 9: Necrotic myofibres (arrows) from a pig subjected to blunt trauma. Bar=500 μ m



FIG 7: Subcutaneous haemorrhage and slight infiltration of inflammatory cells in a pig subjected to blunt trauma. The tissue was frozen before the forensic investigation. H, haemorrhage. Bar=100 μ m

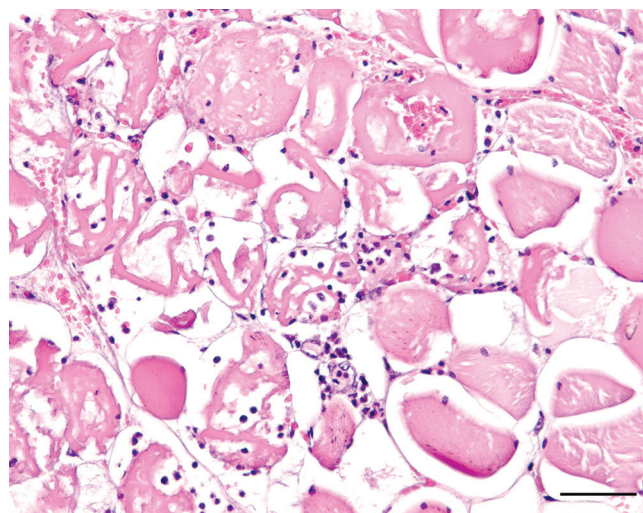


FIG 10: Haemorrhage and necrotic myofibres infiltrated by neutrophils and macrophages from a pig subjected to blunt trauma. Bar=100 μ m

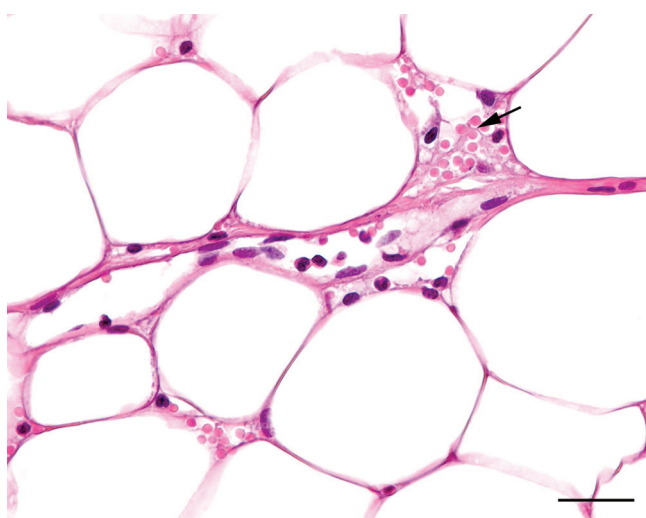


FIG 8: Subcutaneous haemorrhage and infiltration of neutrophils and macrophages in a pig subjected to blunt trauma (arrow=erythrocytes). The tissue was not frozen before the forensic investigation. Bar=50 μ m

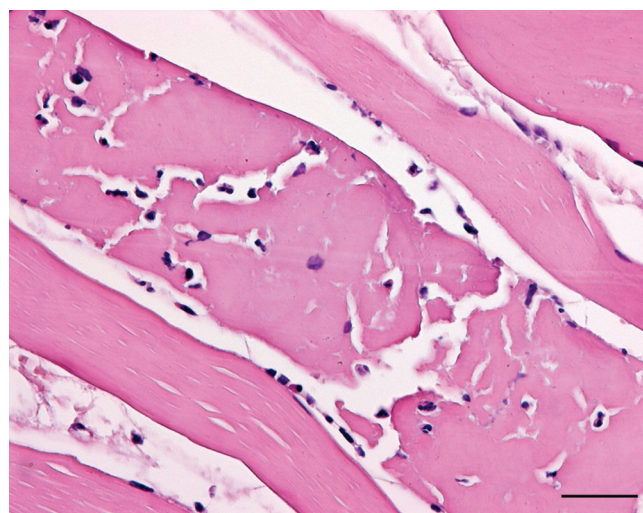


FIG 11: A necrotic myofibre infiltrated by neutrophils from a pig subjected to blunt trauma. Bar=50 μ m

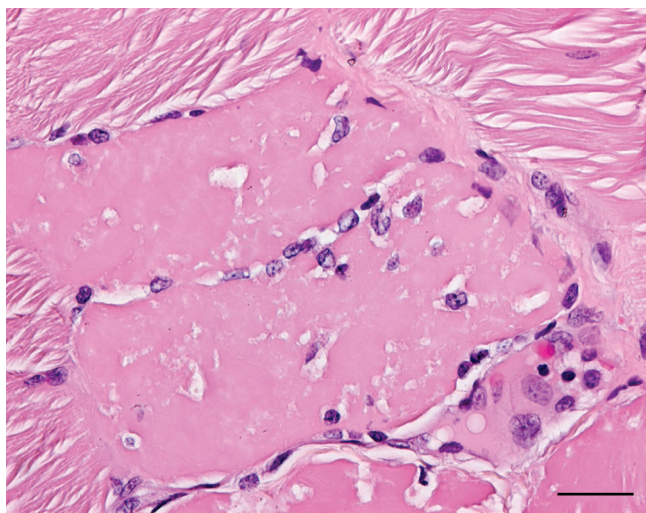


FIG 12: Necrotic myofibres infiltrated by macrophages from a pig subjected to blunt trauma. Bar=50 μ m

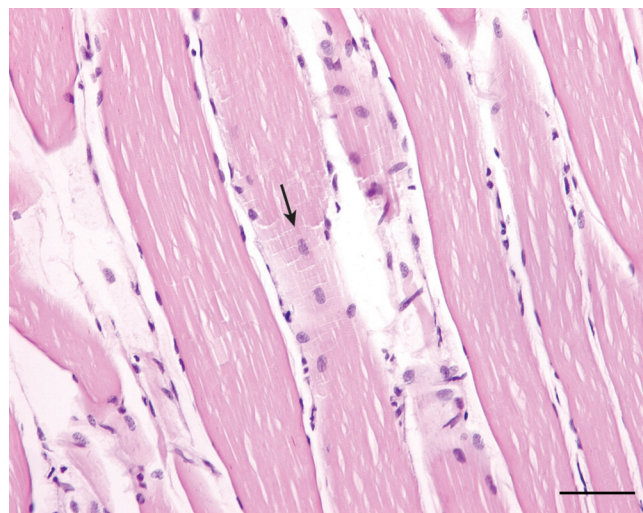


FIG 13: A regenerating myofibre (arrow) with central nuclei from a pig subjected to blunt trauma. Bar=100 μ m

a variable extent, and the neutrophil to macrophage ratio also varied. Extravasated erythrocytes were sometimes present in the interstitial space of the muscles (Fig 10). Rarely, regenerating myofibres with central prominent nuclei (satellite cells) were present (Fig 13).

A written statement on the forensic investigation was available in 82 cases. Where an estimation of age was given ($n=64$), in 81.3 per cent of cases the ages were stated in intervals of one to two hours, ie, two to four hours, four to six hours, and six to eight hours. In the remaining cases, the intervals were stated at between three and 10 hours, ie, two to five hours, four to eight hours, six to 12 hours, nine to 12 hours, and six to 16 hours. In a single case, an interval of one to two days was stated. In 90.6 per cent of the cases the age of the bruises was eight hours or less. The estimations of the age of the bruises were in all cases based on the amount and type of inflammatory cells (Wang and others 2000, 2001, Randeberg and others 2007). In 22.0 per cent of the cases with a written report, the age of the bruises was not determined due to the absence of an inflammatory response. In half of these cases ($n=9$), only samples of skin (no muscle tissue) were received for investigation.

Discussion

Inflicting blunt trauma that causes a bruise in an animal is considered maltreatment and is reported to the police by the Danish Veterinary and Food Administration (Fødevarestyrelsen 2013). Even though the number of cases concerning bruises in pigs has decreased during the last three years, the number of pigs exposed to blunt trauma is still unacceptably high. The decline in the numbers of cases since 2009 presumably is due to an increased focus on the issue, raised both by the industry and the public media. Finally, the Danish Veterinary and Food Administration has established specific registration of bruises caused by blunt trauma in slaughter animals.

Skin bruises in living pigs, if visible, appear vaguely due to the thick epidermis of the porcine skin. The processing of the carcase during slaughter removes the epidermal layer making the bruises easy to identify at postmortem inspection. The gross appearance of bruises found in pigs is similar to bruises in humans caused by blunt trauma. Moreover, in both humans and pigs the pattern of a bruise resembles the object used to inflict the trauma (Saukko and Knight 2004, Armstrong 2005). Impact from sticks such as broom handles and wood or metal rods causes bruises with a tram-line or railway appearance. The bruise consists of two parallel lines of haemorrhage with an undamaged zone in the centre. In a case report by Armstrong (2005), bruises with a tram-line appearance were observed in a 27-year-old man beaten with an expandable baton. Moreover, 'stick-markings' in cattle have been reported (McNally and Warriss 1996). The tram-line appearance of the bruise is caused by stretching and tearing of the vessels nearest to the edges of the object, while the skin perpendicular to the object is compressed but the vessels remain intact (Saukko and Knight 2004).

Sampling of tissue before freezing improves the quality of the histological evaluation, making it especially easier to recognise the extravasated erythrocytes (Figs 7 and 8).

The estimation of the age of bruises in pigs is generally based on studies of wound healing. According to Raekallio (1980) and Saukko and Knight (2004), wounds aged less than four hours are characterised by no or few extravascular neutrophils. Wounds aged four to 12 hours are dominated by neutrophils together with a variable but increasing infiltration of mononuclear cells. Healing of experimental wounds in pigs has been described (Rigal and others 1991, Wang and others 2000, 2001, Gallant and others 2004). Unfortunately, in these models, wounds were studied at the earliest one day after the wounds were created. Randeberg and others (2007) studied experimentally inflicted bruises in pigs and observed large numbers of neutrophils around capillaries and in fatty tissue of the skin after four-and-a-half hours. In addition, Randeberg and others (2007) found that a haemorrhage in the upper skin layers became visible within one to two minutes after a blunt trauma was inflicted. Based on current knowledge of the reaction in bruises, it is possible to estimate the age of bruises in pigs to be more or less than four hours. In bruises more than four hours old, an estimation of age can be proposed based on the pattern of cell infiltration and by comparing the reaction to wound healing, but there will be some uncertainty (Raekallio 1973, Oehmichen 2004).

The fact that 90.6 per cent of bruises in pigs were estimated to be aged eight hours or less showed that the majority of bruises were inflicted shortly before slaughter, during which time the pigs were handled by farmers, drivers and employees at the slaughterhouse. The lack of scientific evidence to determine the precise age of bruises less than four hours old makes it impossible in many cases (41.1 per cent) to determine whose custody the animals were in when the bruises were inflicted. Moreover, it is impossible to estimate the age of bruises in the absence of an inflammatory reaction. Postmortem examinations of three children with bruises known to be at least 30 hours old revealed in some cases an extravasation of erythrocytes in the subcutis but no infiltration of leucocytes, while other bruises in the children exhibited infiltrating inflammatory cells (Byard and others 2008). An extravasation of erythrocytes in the skin and muscle without an accompanying inflammatory response is therefore not necessarily indicative of a bruise inflicted recently. In half of the cases, where an estimation of age was impossible, only skin was sent for forensic investigation. Since the inflammatory reaction was often more advanced in the muscle tissue it is possible that an estimation of age could have been made if muscle tissue had been included in these cases.

In a practice setting, it is mandatory to record the exact localisation and characteristics (uniformity) of lesions and to document these by photos. Moreover, it is mandatory that a histopathological evaluation of the affected skin and muscle is also carried out.

Determining the age of bruises with great certainty is crucial in apportioning blame in a legal context. Visual assessment of bruises as a method to determine the age of a bruise has been shown to be unreliable (Maguire and others 2005, Pilling and others 2010, Grossman and others 2011). Some studies have investigated the value of methods such as spectrophotometry and reflectance spectroscopy, immunohistochemistry, real-time quantitative (RT-q) PCR measurements of mRNA and in situ hybridisation in estimating the age of bruises in both humans and animal models (Takamiya and others 2005, Nakajima and others 2006, Randeberg and others 2007, Hughes and Langlois 2011). Only one of these studies concerns experimental bruises in pigs (Randeberg and others 2007). Therefore, the various methods studied need to be validated on porcine skin before they can assist in the estimation of the age of bruises in pigs.

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