Introduction

Burn management is challenging for every surgeon because it has high mortality, volume and risk. Research and experimental study is needed to create a new management, a new method of burn care. Animals are often used in experimental studies rather than humans, as using humans is more often considered unethical. Therefore, this study was conducted with animals. Animals often used in experimental studies are swine/pigs, guinea pigs, rats/mice, dogs, rabbits and sheep. Porcine models are more frequently used as experimental models because pig skin is anatomically and physiologically more similar to human skin. The cornified layer and epidermis, dermis and subcutaneous region of the pig is similar to human skin. The hair and tubular apocrine of the pig is also similar to that found in humans. Other similarities between porcine and human skin include epidermal enzyme patterns, epidermal tissue turnover time, the character of keratinous proteins, and the composition of the lipid film of the skin surface. Sullivan et al. reported that the porcine model is an excellent tool for the evaluation of therapeutic agents destined to be used in human wounds.

For these reasons, the porcine model is a suitable burn model. It is useful for studying burn symptoms and treatment, such as dressing, tangential excision, skin grafting, scar formation, wound healing, diagnostic tools, fluid resuscitation, and others.

1 Corresponding author: Aditya Wardhana, Diponegoro Road no.71, Jakarta, Indonesia. Tel.: +62 213146938, email: aditya_wrdn@yahoo.com
etc. There is still no gold standard for the procedure to make burn models. Therefore our aim is to conduct a systematic review of how second and third degree burns in porcine models can be used in further studies.

Methods

Types of study
We included systematic reviews, meta analyses and experimental studies of burns that used porcine models for their investigations. Two reviewers identified titles and abstracts of the studies to be included. The full text paper was acquired when there was any uncertainty about its inclusion. We excluded studies that were more than 10 years old and included only thermal injuries.

Types of participant
Porcine model, pig model and swine models that underwent a burning procedure.

Types of intervention
The grading of burn injury had to be second or third degree burns. The grading had to be confirmed using a procedure.

Types of outcome measures
Characteristics of the porcine models (age, weight, kind), area of burn, tool, sources of heat, temperature, temperature measurement device, duration, dimension, validation method and dressing.

Table I - Methodological quality assessment checklist

<table>
<thead>
<tr>
<th>Code</th>
<th>Items</th>
<th>Scores</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Q1</td>
<td>1. Is the hypothesis/aim/objective of the study clearly described?</td>
<td>1 = Yes</td>
<td>If the main outcomes are first mentioned in the Results section, the question should be answered no.</td>
</tr>
<tr>
<td>Q2</td>
<td>2. Are the main outcomes to be measured clearly described in the Introduction or Methods section?</td>
<td>1 = Yes</td>
<td>In cohort studies and trials, inclusion and/or exclusion criteria should be given. In case-control studies, a case-definition and the source for controls should be given.</td>
</tr>
<tr>
<td>Q3</td>
<td>3. Are the characteristics of the patients included in the study clearly described?</td>
<td>1 = Yes</td>
<td>Treatments and placebo (where relevant) that are to be compared should be clearly described.</td>
</tr>
<tr>
<td>Q4</td>
<td>4. Are the interventions of interest clearly described?</td>
<td>1 = Yes</td>
<td>A list of principal confounders is provided.</td>
</tr>
<tr>
<td>Q5</td>
<td>5. Are the distributions of principal confounders in each group of subjects to be compared clearly described?</td>
<td>2 = Yes</td>
<td>Simple outcome data (including denominators and numerators) should be reported for all major findings so that the reader can check the major analyses and conclusions. (This question does not cover statistical tests, which are considered below).</td>
</tr>
<tr>
<td>Q6</td>
<td>6. Are the main findings of the study clearly described?</td>
<td>1 = Yes</td>
<td>In non-normally distributed data the inter-quartile range of results should be reported. In normally distributed data the standard error, standard deviation or confidence intervals should be reported. If the distribution of the data is not described, it must be assumed that the estimates used were appropriate and the question should be answered yes.</td>
</tr>
<tr>
<td>Q7</td>
<td>7. Does the study provide estimates of the random variability in the data for the main outcomes?</td>
<td>1 = Yes</td>
<td>This should be answered yes if the study demonstrates that there was a comprehensive attempt to measure adverse events. (A list of possible adverse events is provided).</td>
</tr>
</tbody>
</table>

Quality assessments tools

To assess the methodological quality of the studies, we used a checklist for the assessment of the methodological quality both of randomized and non-randomized studies of health care interventions. This tool consists of 27 aspects of trial validity and the scoring of them. It is shown in Table I.
| Q9  | Have the characteristics of patients lost to follow-up been described? | 1 = Yes  
0 = No  
0 = Unable to determine | This should be answered yes where there were no losses to follow-up or where losses to follow-up were so small that findings would be unaffected by their inclusion. This should be answered no where a study does not report the number of patients lost to follow-up. |
| Q10 | Have actual probability values been reported (e.g. 0.035 rather than <0.05) for the main outcomes except where the probability value is less than 0.001? | 1 = Yes  
0 = No  
0 = Unable to determine | |

**External validity**

| Q11 | Were the subjects asked to participate in the study representative of the entire population from which they were recruited? | 1 = Yes  
0 = No  
0 = Unable to determine | The study must identify the source population for patients and describe how the patients were selected. Patients would be representative if they comprised the entire source population, an unselected sample of consecutive patients, or a random sample. Random sampling is only feasible where a list of all members are relevant. |
| Q12 | Were those subjects who were prepared to participate representative of the entire population from which they were recruited? | 1 = Yes  
0 = No  
0 = Unable to determine | The proportion of those asked who agreed should be stated. Validation that the sample was representative would include demonstrating that the distribution of the main confounding factors was the same in the study sample and the source population. |
| Q13 | Were the staff, places and facilities where the patients were treated representative of the treatment the majority of patients receive? | 1 = Yes  
0 = No  
0 = Unable to determine | For the question to be answered yes, the study should demonstrate that the intervention was representative of that in use in the source population. The question should be answered no if, for example, the intervention was undertaken in a specialist centre unrepresentative of the hospitals most of the source population would attend. |

**Internal validity – bias**

| Q14 | Was an attempt made to blind study subjects to the intervention they have received? | 1 = Yes  
0 = No  
0 = Unable to determine | For studies where the patients would have no way of knowing which intervention they received, this should be answered yes. |
| Q15 | Was an attempt made to blind those measuring the main outcomes of the intervention? | 1 = Yes  
0 = No  
0 = Unable to determine | |
| Q16 | If any of the results of the study were based on “data dredging”, was this made clear? | 1 = Yes  
0 = No  
0 = Unable to determine | Any analyses that had not been planned at the outset of the study should be clearly indicated. If no retrospective unplanned subgroup analyses were reported, then answer yes. |
| Q17 | In trials and cohort studies, do the analyses adjust for different lengths of follow-up of patients, or in case-control studies, is the time period between the intervention and outcome the same for cases and controls? | 1 = Yes  
0 = No  
0 = Unable to determine | Where follow-up was the same for all study patients the answer should be yes. If different lengths of follow-up were adjusted for by, for example, survival analysis the answer should be yes. Studies where differences in follow-up are ignored should be answered no. |
| Q18 | Were the statistical tests used to assess the main outcomes appropriate? | 1 = Yes  
0 = No  
0 = Unable to determine | The statistical techniques used must be appropriate to the data. For example non-parametric methods should be used for small sample sizes. Where little statistical analysis has been undertaken but where there is no evidence of bias, the question should be answered yes. If the distribution of the data (normal or not) is not described it must be assumed that the estimates used were appropriate and the question should be answered yes. |
| Q19 | Was compliance with the intervention/s reliable? | 1 = Yes  
0 = No  
0 = Unable to determine | Where there was non compliance with the allocated treatment or where there was contamination of one group, the question should be answered no. For studies where the effect of any misclassification was likely to bias any association to the null, the question should be answered yes. |
| Q20 | Were the main outcome measures used accurate (valid and reliable)? | 1 = Yes  
0 = No  
0 = Unable to determine | For studies where the outcome measures are clearly described, the question should be answered yes. For studies which refer to other work or that demonstrate the outcome measures are accurate, the question should be answered yes. |

**Internal validity - confounding (selection bias)**

| Q21 | Were the patients in different intervention groups (trials and cohort studies) or were the cases and controls (case-control studies) recruited from the same population? | 1 = Yes  
0 = No  
0 = Unable to determine | For example, patients for all comparison groups should be selected from the same hospital. The question should be answered unable to determine for cohort and case-control studies where there is no information concerning the source of patients included in the study. |
### Results

**Description of studies**

A search of Ovid MEDLINE(R) database (http://ovidsp.ovid.com) from the year 2006 until 2017 revealed 12 studies; the search of the PubMed database resulted in 28 studies from 2006 until 2017; 108 studies between 2006 and 2017 were retrieved from the Burns Journal database, and lastly the COCHRANE LIBRARY search found 2 studies for the period 2006 until 2017. The online search ended on Monday, 6th July 2015 at 4:00 pm. A total of 150 studies were retrieved from 4 databases. We identified and excluded studies based on our inclusion and exclusion criteria, thus yielding 23 remaining studies. Finally, after we had read the abstracts and excluded two based on full text availability, the remaining 21 studies were selected for the final analysis. See Fig. 1 for a detailed search history and Table II for results of the search.

#### Risk of bias among the studies included

The methodological quality assessment is a method to assess both randomized and non-randomized studies. This checklist consists of 5 subscales:

- reporting, to allow a reader to make an unbiased statement of the findings of the study;
- external validity, whether the study could be generalized to the population from which the study subjects were derived;
- bias, assessed biases in the measurement of the intervention and the outcome;
- power, to detect a clinically important difference where the probability value for a difference being due to chance is less than 5%.

<table>
<thead>
<tr>
<th>Question</th>
<th>Score</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Q22</td>
<td>1 = Yes</td>
<td>Were study subjects in different intervention groups (trials and cohort studies) or were the cases and controls (case-control studies) recruited over the same period of time?</td>
</tr>
<tr>
<td>Q23</td>
<td>1 = Yes</td>
<td>Were study subjects randomized to intervention groups?</td>
</tr>
<tr>
<td>Q24</td>
<td>1 = Yes</td>
<td>Was the randomized intervention assignment concealed from both patients and health care staff until recruitment was complete and irrevocable?</td>
</tr>
<tr>
<td>Q25</td>
<td>1 = Yes</td>
<td>Was there adequate adjustment for confounding in the analyses from which the main findings were drawn?</td>
</tr>
<tr>
<td>Q26</td>
<td>1 = Yes</td>
<td>Were losses of patients to follow-up taken into account?</td>
</tr>
<tr>
<td>Q27</td>
<td>&lt;n1 = 0</td>
<td>Did the study have sufficient power to detect a clinically important effect where the probability value for a difference being due to chance is less than 5%?</td>
</tr>
</tbody>
</table>

For a study that does not specify the time period over which patients were recruited, the question should be answered as unable to determine.

Studies that state that subjects were randomized should be answered yes except where method of randomization would not ensure random allocation. For example, alternate allocation would score no because it is predictable.

All non-randomized studies should be answered no. If assignment was concealed from patients but not from staff, it should be answered no.

This question should be answered no for trials if: the main conclusions of the study were based on analyses of treatment rather than intention to treat; the distribution of known confounders in the different treatment groups was not described; or the distribution of known confounders differed between the treatment groups but was not taken into account in the analyses. In non-randomized studies if the effect of the main confounders was not investigated or confounding was demonstrated but no adjustment was made in the final analyses, the question should be answered no.

If the numbers of patients lost to follow-up are not reported, the question should be answered as unable to determine. If the proportion lost to follow-up was too small to affect the main findings, the question should be answered yes.

Sample sizes have been calculated to detect a difference of x% and y%.

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**Fig. 1** - Detailed search history diagram.
• confounding, addressed bias in the selection of study subjects;
• power, to assess whether the negative findings from a study could be due to chance.

A summary of methodological quality assessment is presented in Table III.

Table III shows the results of assessment through 27 questions representing 5 subscales that are presented in Table I. Out of the 21 studies assessed, 16 have a good score and 5 have a fair score, with a median of 21 and averaging 20.5. This indicates that most of the studies are in the considerable range, and averaging a good score of assessment, making them methodologically a suitable source for this review.

<table>
<thead>
<tr>
<th>Study ID</th>
<th>Author</th>
<th>Title</th>
<th>Included</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Wang (2008)</td>
<td>“Conservative surgical debridement as a burn treatment: supporting evidence from a porcine burn model”</td>
<td>Included</td>
</tr>
<tr>
<td>2</td>
<td>Singer (2009)</td>
<td>“Healing of mid-dermal burns in a diabetic porcine model”</td>
<td>Included</td>
</tr>
<tr>
<td>3</td>
<td>Singer (2011)</td>
<td>“Reepithelialization of mid-dermal porcine burns after rapid enzymatic debridement with Debrase®”</td>
<td>Included</td>
</tr>
<tr>
<td>4</td>
<td>Singer (2010)</td>
<td>“The effects of rapid enzymatic debridement of deep partial-thickness burns with Debrase® on wound reepithelialization in swine.”</td>
<td>Included</td>
</tr>
<tr>
<td>5</td>
<td>Fourman (2014)</td>
<td>“Indocyanine green dye angiography accurately predicts survival in the zone of ischemia in a burn comb model.”</td>
<td>Included</td>
</tr>
<tr>
<td>6</td>
<td>Branski (2011)</td>
<td>“Fibrin sealant improves graft adherence in a porcine full-thickness burn wound model.”</td>
<td>Included</td>
</tr>
<tr>
<td>9</td>
<td>Wang (2009)</td>
<td>“Burn healing is dependent on burn site: A quantitative analysis from a porcine burn model.”</td>
<td>Included</td>
</tr>
<tr>
<td>10</td>
<td>Cancio (2007)</td>
<td>“Intestinal and gastric tonometry during experimental burn shock.”</td>
<td>Included</td>
</tr>
<tr>
<td>15</td>
<td>Li (2015)</td>
<td>“A topical aqueous oxygen emulsion stimulates granulation tissue formation in a porcine second-degree burn wound.”</td>
<td>Included</td>
</tr>
<tr>
<td>16</td>
<td>Sheu (2014)</td>
<td>“The pig as an experimental model for mid-dermal burns research.”</td>
<td>Included</td>
</tr>
<tr>
<td>17</td>
<td>King (2015)</td>
<td>“Surgical wound debridement sequentially characterized in a porcine burn model with multispectral imaging.”</td>
<td>Included</td>
</tr>
<tr>
<td>18</td>
<td>Cuttle (2006)</td>
<td>“A porcine deep dermal partial thickness burn model with hypertrophic scarring.”</td>
<td>Included</td>
</tr>
<tr>
<td>21</td>
<td>Chan (2012)</td>
<td>“The correlation between time to skin grafting and hypertrophic scarring following an acute contact burn in a porcine model.”</td>
<td>Included</td>
</tr>
</tbody>
</table>
Fig. 2 describes the frequency of methodological quality assessment scoring for each study. Twelve of the studies used metals (including aluminium and brass) as a tool of intervention, 11 of which have a good score and only 1 has a fair score. The other tool used for intervention was a glass bottle filled with hot water, in 9 studies in total, 6 of which have a good score and 3 have a fair score.

**Effects of intervention**

The method that resulted in partial thickness burn wounds in the porcine model is presented in Table IV, while the method that resulted in full thickness burn wounds in the porcine model is presented in Table V. The temperature and duration of exposure used to create partial thickness and full thickness wounds is listed in Fig. 3 and Fig. 4, respectively.

The data show that the same number of studies (7) used metal or glass bottle as a tool. The most common heat source was heated water (6), followed by microwave (4) and hot plate (1).
The average exposure temperature to create partial thickness burn wounds was 94.21°C, with a median of 92°C, and the average duration was 19.79s, with a median of 15.5s.

The above data show that the most used tool was metal (4), followed by glass bottle in just one study less (3). There are an equal number of studies that used heated water and gas burner (2), with none using microwave or hot plate. The average exposure temperature to create full thickness burn wounds was 139.14°C, with a median of 100°C, and the average duration was 48.14s, with a median of 30s. Therefore, if we compare both data sets, we see that it takes a higher temperature and a longer duration of exposure to create a full thickness burn wound in the porcine model, ignoring the heat source and tools used.

The porcine models weighed 15-65 kg. Female and male pigs were used. Not every study described the age of the pigs. The age range was 6-42 weeks. Out of 22 studies, 18 made the burn injury on the back and flank of the pig’s body. Prior to any surgical intervention, the hair on the back and flank was clipped with an electric clipper and the skin was rinsed sequentially with an antiseptic solution and clean water.

Three methods were used to create full thickness burn wounds: aluminium, brass and immersion in hot water. Regarding the aluminium method, an aluminium bar was heated to 200°C and placed on the pig’s skin for 20 seconds. As for the brass comb/bar method, the brass was heated to 100°C and placed on the pig’s skin for 20-30 seconds.

The tools used to make the burn wound were mostly heated in boiling water to the desired temperature. Regarding immersion in hot water, the water was heated to 70-97°C and
the pigs were immersed in the water for 17-180 seconds.

Various methods were used to create deep dermal burns, the most frequent being a modified glass bottle. The bottom of the bottle-heated device was removed and replaced with a plastic wrap. Then the bottle was filled with sterile water and heated to the desired temperature (92°C). The bottle was placed on the pig’s skin for 15 seconds. The other method used an aluminium bar heated to 80°C and then placed on the pig’s skin for 20 seconds with gravity pressure.

To measure the depth of the burn wound, almost all of the studies used histopathological evaluation. This evaluation was achieved from biopsies done periodically. Singer et al. did the biopsies at 7, 9, 11 and 13 days after injury. Tennyson et al. used Histologic Assessment Tools, which consists in 8 tissue measurements.

**Discussion**

A burn wound model should have proper extent and depth of wound. However, the burn wound model still has to be safe, simple and reproducible. The important elements needed to produce a burn wound model are the tools, temperature, duration of exposure and pressure. Animal models should have similar characteristics to human skin and have a similar outcome of exposure and pressure. Animal models should have similar type and extent of burn when exposure is applied.2,25

From our perspective, the safest, simplest and most reproducible tool for creating deep dermal burns is the modified glass bottle. It is safe because we can monitor burn appearance during creation from the bottom of the bottle. However, there are no studies that compare the modified glass bottle with the metal bar to make full thickness burn wounds. To our knowledge, they are caused by the better conduction of metal compared to glass. To date, the metal bar (aluminium and brass) is still used to make full thickness burn wounds in the porcine model. The other technique is to immerse the porcine models in hot water, but this is ethically harder to accept. The weaknesses of the studies are the small sample sizes and the fact the sample cannot be randomized.

**Conclusion**

A review of the studies above showed that there was no standardized method to create burn wounds in the porcine model. Nevertheless, for deep dermal burn wounds we can use the modified glass bottle method and for full thickness burn wounds we can use metal tools (aluminium or brass). There are no previous studies that discuss how to make porcine burn models. There are also no studies in this review that focus on creating the burn wound alone. Further studies are needed to achieve better results in how to create burn wounds in porcine models.

**BIBLIOGRAPHY**


