

*Original Article*

## Modified Thiel and saturated salt solutions for rabbit soft cadaveric embalming: A preliminary study

Jirawat Saetan<sup>1\*</sup>, Jongdee Nopparat<sup>1, 2</sup>, Chittipong Tipbunjong<sup>1</sup>, Wipapan Khimmaktong<sup>1</sup>,  
Uraporn Vongvatcharanon<sup>1</sup>, Kosol Puriwattanakul<sup>1</sup>, Boonsin Tangtrakulwanich<sup>3</sup>,  
Chongdee Thammakhet-Buranachai<sup>2, 4</sup>, and Wandee Udomuksorn<sup>1</sup>

<sup>1</sup> Division of Health and Applied Sciences, Faculty of Science,  
Prince of Songkla University, Hat Yai, Songkhla, 90110 Thailand

<sup>2</sup> Center of Excellence for Trace Analysis and Biosensor,  
Prince of Songkla University, Hat Yai, Songkhla, 90110 Thailand

<sup>3</sup> Department of Orthopedics, Faculty of Medicine,  
Prince of Songkla University, Hat Yai, Songkhla, 90110 Thailand

<sup>4</sup> Division of Physical Science, Faculty of Science,  
Prince of Songkla University, Hat Yai, Songkhla, 90110 Thailand

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### Abstract

Soft cadaveric embalming is beneficial for gross anatomy practice as well as for its low formaldehyde content. Examples of soft cadaveric embalming fluids are the Thiel solution (TS) and the newly developed saturated salt solution (SSS). In this study, we descriptively demonstrated the effects of TS, SSS, and conventional formaldehyde-based solution (FAS) on gross morphology, histology, range of motion (ROM) of joints, microbial growth and satisfactory level of five rabbit bodies. The TS embalmed rabbits demonstrated both muscular and tendon fragmentation while the SSS embalmed rabbit presented only muscular fragmentation. This finding indicated that, in order to gain the softness of tissues mimicking the *in vivo* conditions, these fluids could directly act through muscle and tendon fibers. By ROM measurement, the TS and SSS rabbit had greater joint flexibility than the FAS rabbit. Slight microbial growth on TS and SSS embalmed rabbit was detected. User satisfaction on various tested parameters was also reported.

**Keywords:** soft cadavers, Thiel solution, saturated salt solution, rabbits, anatomy

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### 1. Introduction

Anatomy teaching relies on the use of cadaver. Advantages of cadaver are discussed in many aspects, for example, the three-dimensional learning of the human body (McLachlan, Bligh, Bradley & Searle, 2004). Attenuation of cadaver dissection is bound to the impaired scientific ability of medical student during the diagnosis process (Aziz *et al.*, 2002; Aziz & McKenzie, 1999). To preserve the cadaver, the

embalming fluid is crucial. Good embalming fluid must retain the natural color of structures; prevent dehydration and microorganism growth; and be the least toxic as possible (Coleman & Kogan, 1998). There are three main ingredients that make up the embalming fluid including preservatives, disinfectants, and modifying agents (Brenner, 2014).

The commonly used preservatives are formaldehyde ( $\text{CH}_2\text{O}$ ), glutaraldehyde, and various forms of alcohol (Brenner, 2014). Sodium borate is the component used in Thiel embalming fluid, the well-known low formaldehyde fluid for soft cadaver embalming (Benkhadra *et al.*, 2011; Macdonald & MacGregor, 1997; Thiel, 1992). The second component is disinfectants which are often added to the

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\*Corresponding author  
Email address: [jisaetan@gmail.com](mailto:jisaetan@gmail.com)

embalming solutions in order to limit the fungal, bacterial, and viral growth as well as to prevent infectious agents transmitted from cadavers to the embalmers. The disinfectants include phenol and its derivatives which destroy cell wall, thus causing protein precipitations in the microbe cells (Bedino, 1994). Many embalming formulas contain various phenolic compounds, e.g., phenol, salicylic acid (2-carboxyphenol), thymol (2-isopropyl-5-methylphenol), and sodium pentachlorophenate. Lastly, the modifying agents are mainly buffers with suggested pH at 7.38-7.40. The sodium borate, sodium bicarbonate, sodium carbonate, and magnesium carbonate buffers are widely employed (Brenner, 2014). Salt solution such as calcium chloride, potassium thiocyanide, and ammonium sulphate are also counted as modifying agent. Besides buffer and salt, glycerine, chloral hydrate, mono-, di-, and polyethylene glycol, sorbital, sodium lauryl sulphate, and sodium 2-sulphonatoethyl laurate are also used (Brenner, 2014; Schep, Slaughter & Temple, 2009).

Although there are various components in the embalming fluids, they are mainly classified into two types based on the amount of formaldehyde. The formaldehyde-based formulas contain primarily formaldehyde and other modifying agents such as Karlsbad salts, chloral hydrate, alcohol, glycerine, phenol, sodium arsenate, and salicylic acid, 6-chlorothymol (Brenner, 2014). This type holds good preservation properties but usually complicates the handling with great rigidity, joint stiffness, color change, and dehydration of cadavers (Brenner, 2014; Hayashi *et al.*, 2014; Healy *et al.*, 2015; Hunter, Eisma & Lamb, 2014). Importantly, their carcinogenic properties are a matter of concern. Occasional exposure of formalin can cause eye, nose, throat, and respiratory tract irritations while prolonged exposure leads to headache, dizziness, and genetic damages (Frolich, Andersen, Knutsen, & Flood, 1984; Mirabelli, Holt & Cope, 2011). In contrast, the newly introduced low- to non-formaldehyde-based formulas claim better outcomes such as softness and flexibility of the cadaver. The use of ionic salt as preservative was first suggested by Coleman and Kogan (1998). Recently, the soft embalming method namely Thiel embalming solution has become widely used (Hunter *et al.*, 2014; Sangchay, 2014; Thiel, 1992). This solution relies mainly on salt, ethylene glycol, and phenol. With this solution many benefits are earned compared to the conventional formalin fluids as it provides high satisfaction scores on the cadaveric odor, and muscular identification and quality (Sangchay, 2014). Thiel-embalmed cadavers are predominantly reported for the great range of motion of joints (Balta *et al.*, 2019; Hayashi *et al.*, 2014), tissue elasticity, pliability and separation of planes, red muscle color, realistic adipose tissue color, and joint flexibility (Healy *et al.*, 2015; Liao, Kemp, Corner, Eisma & Huang, 2015). Hence, the Thiel fluid benefits several surgical trainings and practices (Cabello *et al.*, 2014; Eisma, Lamb & Soames, 2011; Healy *et al.*, 2015; Sangchay, 2014). However, the high costs to produce this solution must be considered (Hammer *et al.*, 2015). Hayashi *et al.* (2014) reported the saturated salt solution method (SSS), which is a cheaper embalming fluid and is feasible for surgical skills training. The SSS is primarily based on sodium chloride (NaCl) while the other components are like those present in conventional formalin solution. However, the percentage of formalin is as low as seen in Thiel solution. The SSS-embalmed cadavers show no signs of microbial

infections but have less joint flexibility and elasticity compared to Thiel-embalmed cadaver; nevertheless this newly method reaches equal satisfaction scores evaluated by the surgeons (Hayashi *et al.*, 2014). Moreover, the SSS-embalmed cadavers are reported to retain realistic soft skin resembling that in the live patient, which is suitable for many flap surgeries (Shirai, Hayashi, & Itoh, 2015).

At present, the Prince of Songkla University (PSU) Cadaveric Surgical Training Center is established and held by Department of Anatomy, Faculty of Science and Department of Orthopedic Surgery and Physical Medicine, Faculty of Medicine, PSU. However, this center runs surgical training largely based on fresh cadaver which requires a significant amount of preparation time for freezing and defrosting the cadaver. Hence, in an attempt to develop soft cadaver to serve the cadaveric center as well as anatomy classes at PSU, this study was aimed to preliminarily demonstrate and compare the embalming efficiency of Thiel solution and SSS in rabbits.

## 2. Materials and Methods

### 2.1 Experimental design

All rabbits (n=5) used in this study were under the approval of the animal ethic committee, Prince of Songkla University (MOE 0521.11/926). The formulas of the embalming solutions are shown in Table 1 (Hayashi *et al.*, 2014). The rabbits embalmed by TS were named TS(A) and TS(B) while those embalmed by SSS were SSS(A) and SSS(B). Also, one rabbit embalmed by conventional formaldehyde solution was named FAS. The rabbits were euthanized with a lethal dose of thiopental (70 mg/body weight) and immediately embalmed with two to three liters of the mentioned solutions. The injection route was either through central ear artery or cardiac puncture where the solution was loaded into the rabbit body by gravitation. After 2 days, the fully embalmed rabbits were placed in a sealed plastic bag and stored at room temperature for 3 months. The morphology, as well as other parameters, was investigated after 3 months of embalming (Hayashi *et al.*, 2014).

### 2.2 Histological studies of muscular and tendon tissues

The rigidity of muscles and tendons is one of the major drawbacks of the conventional formaldehyde embalmed method. Hence, in order to demonstrate the effects of embalming fluids, the adductor muscles and Achilles tendon from all rabbits were dissected and routinely processed through paraffin technique. Briefly, the seven micron-thick sections were cut, put onto glass slides, deparaffinized in xylene, hydrated through graded ethanol to distilled water, and finally stained with either Hematoxylin and Eosin (H&E) or Masson's trichrome techniques. Images were captured by a Nikon E600 microscope, with a digital camera (Nikon, Tokyo, Japan). Fragmentation at the cellular level was investigated (Benkhadra *et al.*, 2011).

### 2.3 Measurement of range of motion (ROM) of joints

The rabbit bodies were positioned supine. The shoulder, elbow, wrist, hip, knee, and ankle joints were

Table 1. Chemical formulas for rabbit embalming; FAS: formaldehyde solution, TS: Thiel's solution (Hayashi *et al.*, 2014); SSS: saturated salt solution (Hayashi *et al.*, 2014)

FAS		TS		SSS	
Solution	Amounts	Solution	Amounts	Solution	Amounts
Stem A		Stem A		Sodium chloride	20 kg (saturated)
Potassium nitrate	0.5 kg	4-chloro-3-methylphenol	66 g	20% formaldehyde	1 L
Water	9 L	Propylene glycol	0.66 L	Phenol	0.2 L
Heat until the volume is	6 L	Stem B		Glycerine	0.5 L
Stem B		Ammonium nitrate	2500 g	Isopropyl alcohol	4 L
Glycerine	2 L	Hot water	4 L	Water	19.3 L
Methanol	4 L	Stem C		Total	25 L
Phenol	0.4 L	Boric acid	370 g		
37-40% formaldehyde	2.5 L	Potassium nitrate	620 g		
Final solution		Hot water	5 L		
Stem A	6 L	Stock solution			
Stem B	8.4 L	Stem A	0.66 L		
Total	14.4 L	Stem B	4 L		
		Stem C	5 L		
		Propylene glycol	3.7 L		
		Hot water	3.3 L		
		Final solution			
		Stock solution	16.66 L		
		Sodium sulfite	800 g		
		20% formaldehyde	0.6 L		
		Morpholine	0.3 L		
		Ethanol	1.3 L		
		Total	18.86 L		

subjected for passive ROM measurements by two main joint motions; flexion and extension. The full extent of joint mobility was measured into a degree of movement using goniometer (Soucie *et al.*, 2011).

## 2.4 Microbial growth determination

The growth of bacteria, mold, and yeast of four different regions of rabbit bodies; the pharynx, pleural cavity, abdominal cavity, and rectum were evaluated (Hayashi *et al.*, 2014). This test was performed and evaluated by a laboratory technician from Department of Microbiology, Faculty of Science, PSU. Briefly, the samples from each area were collected by swabbing technique and dispersed in phosphate buffered saline. The cell suspension was then diluted in a series of a 10-fold dilutions in order to obtain plates with a countable number of microbes. A 100  $\mu$ l from each dilution was plated in replicates of three on petri dishes with nutrient agar intended for the bacteria, mold, and yeast. The petri dishes were then incubated in suitable environments and for an appropriate time before microbial colonies were counted. The amount of microbial growth was expressed as colony-forming unit (CFU)/point.

## 2.5 Satisfaction level determination

Satisfaction levels of all rabbit bodies were assessed by questionnaire evaluation and scoring. The checked parameters included the cadaveric odor, visual and tactile assessments, muscle and tendon consistency, joint mobility, nerve and vessel appearances and qualities, and subcutaneous fat and skin quality. Muscle and tendon consistency can be defined by their post-embalming textures, which closely

resemble to *in vivo* conditions (Sangchay, 2014). The questionnaires were completed by graduate students, scientists, and researchers of the Department of Anatomy as well as by second year medical students of PSU.

## 2.6 Statistical analysis

All evaluated scores reflecting satisfaction levels were expressed individually and in terms of mean  $\pm$  SD. All data were tested for homogeneity of variances. The One-way ANOVA followed by either Bonferroni's or Tamhane's post hoc analysis was used to test the level of mean differences among five bodies and significant different level designated if the p-value was less than 0.05.

## 3. Results

### 3.1 Gross morphology

Through observation by naked eye, the adductor muscle integrity of FAS (Figure 1A-B) closely resembled natural tissue in comparison with TS- and SSS-embalmed rabbits (Figure 1C-F). The color of adductor muscles in all rabbits was intensely brown, red-brown to dark brown. In addition, the FAS-embalmed rabbit was easily distinguished by the femoral vessels and nerve (Figure 1B) while those of TS- and SSS-embalmed rabbits appeared moist and soft (Figure 1D-E).

### 3.2 Histology of skeletal muscle and tendon

Hematoxylin and Eosin (H&E) staining of 7  $\mu$ m thick sections of skeletal muscle and tendon were

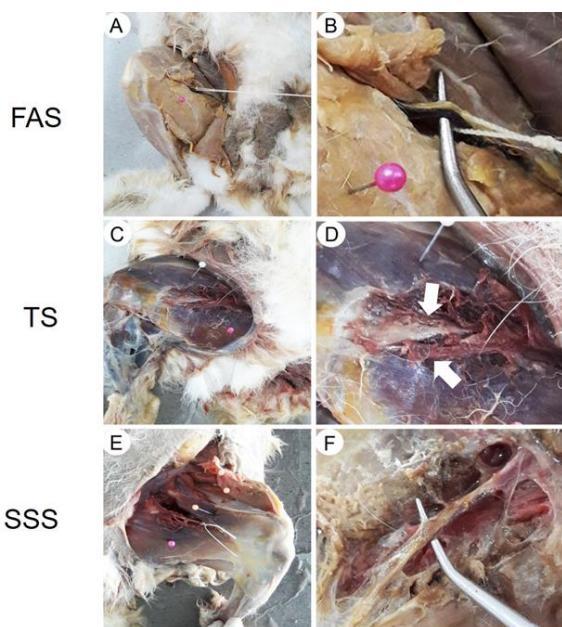


Figure 1. Photographs demonstrating adductor muscles appearance at thigh region of three months embalmed rabbits with different embalming formula. A-B FAS, C-D TS, and D-F SSS. The femoral nerve and artery were placed down on the probe or pointed by the white arrows.

demonstrated. The results revealed that the skeletal muscle architecture of the TS and SSS groups were fragmented (Figure 2D, G, and H), while the muscle fibers of the FAS group were well fixed with noted normal appearance (Figure 2A-B). Muscle fibers of the FAS were normal, in which they had multiple nuclei located at the periphery of the cells, while this characteristic was absent in the TS- and SSS-embalmed rabbits. However, the striation of muscle fiber was still seen in TS and SSS but not in FAS groups (Figure 2B, E, and H). Notably, the fragmentations appearing in muscle fibers of the TS- and SSS-embalmed rabbits were slightly different. Fine fragmentation was detected in the muscle fiber of TS-embalmed rabbits (Figure 2D inset) while coarse fragmentation was obvious in those of SSS-embalmed rabbits (Figure 2G-H arrows). For tendon histology, they exhibited regular dense connective tissue, and no significant difference in collagen fibers arrangement was seen in tendons of FAS and SSS-embalmed rabbits (Figure 2C, I). However, collagen fibers of TS-embalmed rabbits also allowed fragmentation (Figure 2F). Masson's trichrome staining was performed to confirm the collagen fibers arrangement and the results were in accordance with those of H&E staining. Blue staining of the perimysium and endomysium of skeletal muscles was dominant in TS-embalmed rabbits (Figure 3D-E). The tendons of all rabbits were intensely stained (Figure 3C, I) with some level of fragmentation was seen in those of TS-embalmed animals (Figure 3F).

### 3.3 Range of motion (ROM) of joints

The ROM values of five rabbits embalmed by three different methods are shown in Table 2. The ROM of TS(A) and TS(B) exhibited the greatest joint flexibility followed by

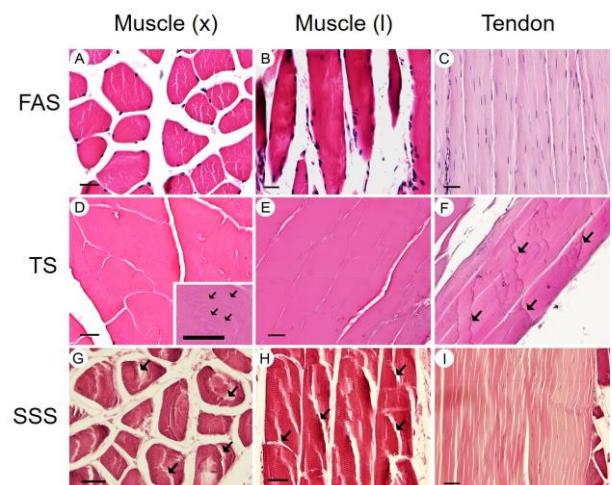


Figure 2. Hematoxylin and Eosin staining of muscle fibers and tendons (x; cross section, l; long section) of the FAS-embalmed rabbit (A-C), TS-embalmed rabbits (D-E) and SSS-embalmed rabbits (G-I). The D inset demonstrated fine fragmentation of muscle fibers. Arrows indicate fragmentations of muscle fibers; Bars: 50  $\mu$ m (A-I), 25  $\mu$ m (D inset).

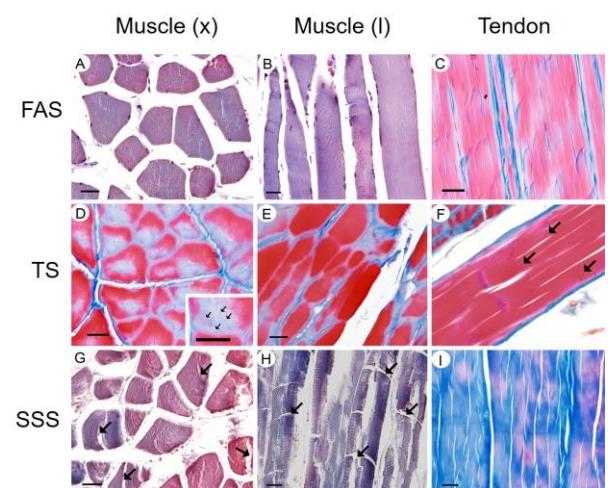


Figure 3. Masson's trichrome staining of skeletal muscles and tendons (x; cross section, l; long section) of the FAS-embalmed rabbit (A-C), TS-embalmed rabbits (D-E) and SSS-embalmed rabbits (G-I). The D inset demonstrated fine fragmentation of muscle fibers. Arrows indicate fragmentations of muscle fibers; Bars: 50  $\mu$ m (A-I), 25  $\mu$ m (D inset).

the ROM of SSS(A) and SSS(B). In this study, the TS and SSS-embalmed rabbits had higher ROM values than those of the FAS-embalmed rabbit. Notably, the ROM values of the shoulder and hip joints of the FAS-embalmed rabbit were zero, reflecting the poorest joint mobility.

### 3.4 Bacterial and Fungal tests

Microbial analysis revealed that the FAS and SSS(B)-embalmed rabbits exhibited low bacterial growth in all tested points (Table 3). Conversely, the TS- and SSS(A)-

Table 2. Range of motion (ROM) of joints at the shoulder, elbow, wrist, hip, knee, and ankle of embalmed rabbits for all three methods including FAS, TS (A and B) and SSS (A and B). Letters in parenthesis refer to the name of the animal.

Joint	Action	FAS		TS(A)		TS(B)		SSS(A)		SSS(B)	
		R	L	R	L	R	L	R	L	R	L
Shoulder	Flexion	0	0	40	28	24	46	22	18	14	10
	Abduction	0	0	88	88	50	80	34	22	34	26
	Extension	0	0	32	32	48	48	20	18	16	18
Elbow	Flexion	26	30	22	44	48	42	14	28	19	16
	Extension	20	22	80	56	62	72	64	30	42	18
Wrist	Flexion	16	18	100	94	88	94	110	20	90	30
	Extension	10	18	56	52	20	44	70	80	70	50
Hip	Flexion	0	0	32	24	38	32	40	70	30	16
	Extension	0	0	30	28	38	14	60	30	42	10
Knee	Flexion	14	20	44	40	68	40	30	50	48	40
	Extension	32	22	56	48	42	58	55	30	38	48
Ankle	Flexion	18	10	52	86	30	56	28	36	26	42
	Extension	30	12	82	70	70	34	42	58	42	32

Table 3. The bacterial and fungal culture tests of five rabbit bodies embalmed with FAS, TS (A and B) and SSS (A and B). Letters in parenthesis refer to the name of the animal.

Region	FAS	TS(A)	TS(B)	SSS(A)	SSS(B)
Pharynx					
Bacteria (CFU/point)	13	$6.9 \times 10^2$	5	$3.9 \times 10^2$	8
Mold and yeast (CFU/point)	0	0	8	10	0
Pleural cavity					
Bacteria (CFU/point)	13	$1.4 \times 10^3$	$1.4 \times 10^3$	33	20
Mold and yeast (CFU/point)	0	0	13	0	0
Abdominal cavity					
Bacteria (CFU/point)	13	$1.4 \times 10^3$	$1.4 \times 10^3$	$1.5 \times 10^2$	3
Mold and yeast (CFU/point)	0	0	3	0	0
Rectum					
Bacteria (CFU/point)	3	$3 \times 10^2$	$7.8 \times 10^2$	$1.0 \times 10^5$	$7 \times 10^2$
Mold and yeast (CFU/point)	0	0	3	0	0

embalmed rabbits presented some bacterial growth for all tested samples. However, their growth was not defined as problematic in anatomy teaching because there were no bacterial colonies seen by the naked eye. The mold and yeast growth were completely restricted in all rabbit samples except those of TS(B), which showed a small number of CFU/point (Table 3). Conclusively, TS- and SSS-embalming methods effectively prevented bacterial and fungal growths whereas the FAS-embalming method exhibited an excellent antiseptic property.

### 3.5 Satisfaction level of users

The satisfaction level on each rabbit body embalmed by each method was blindly evaluated using a 5-point rating scale (1 = completely different, 2 = somewhat different, 3 = neither different nor similar, 4 = somewhat similar, and 5 = completely similar, from living animals). There were 34-37 evaluators for FAS-, SSS(A)- and SSS(B)-embalmed rabbits while only 14 evaluators for TS(A)- and TS(B)-embalmed rabbits. The users felt that the odor of the FAS-embalmed rabbit was significantly stronger than that of the SSS(A)-embalmed rabbit ( $p = 0.015$ ) while score of odors of the TS- and SSS-embalmed rabbits were similar. The score of visual assessment of the TS(B)-embalmed rabbit was

significantly greater than that of the FAS-embalmed rabbit ( $p = 0.032$ ). For the tactile assessments and muscle consistency, no significant difference in mean scores of all rabbits was found. The tendon consistency of the SSS(B)-embalmed rabbit was evaluated to be significantly greater than that of the FAS-embalmed rabbit ( $p = 0.010$ ) while the joint mobility of the TS(B)-embalmed rabbit was significantly more flexible than that of the FAS-embalmed rabbit ( $p = 0.027$ ). The TS(B)-embalmed rabbit also had a higher score of joint flexibility than that of the SSS(B)-embalmed rabbit ( $p = 0.012$ ). Interestingly, the subcutaneous fat and skin quality score of the FAS-embalmed rabbit was significantly lower than those of all SSS- and TS-embalmed rabbits ( $p = 0, 0.002, 0.003$  and  $0.028$ ) and no significant difference in mean score was detected for the test of nerve and vessel appearances and qualities (Figure 4).

### 4. Discussion and Conclusions

In this study, three standard methods; the FAS, TS, and SSS (Hayashi *et al.*, 2014, Sangchay, 2011) were utilized to embalm five rabbits. This study was considered as preliminary work showing the effects of each embalming method on the rabbit bodies. Gross morphology of rabbit muscle from all three methods was dark in color, either dark

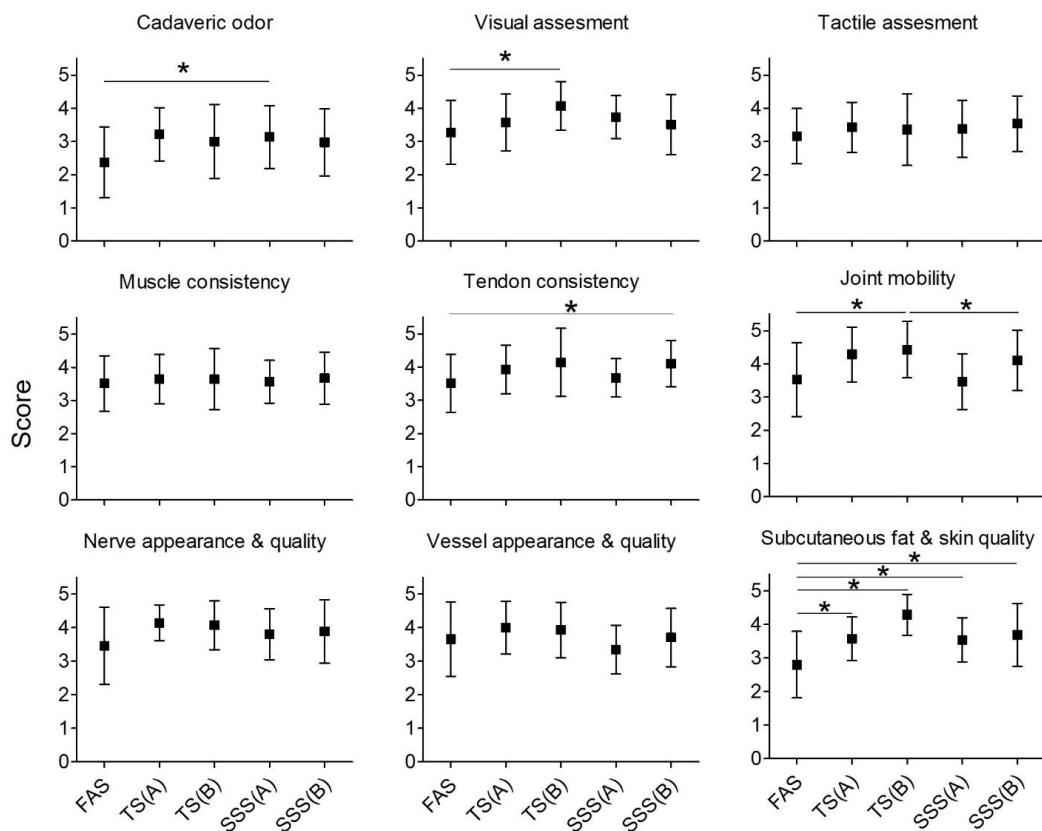


Figure 4. Results showing satisfaction level of rabbit bodies embalmed by three methods. Evaluated parameters included cadaveric odor, visual assessment, tactile assessment and muscle consistency, tendon and joint consistencies, joint mobility, nerve and vessel appearances and qualities, and subcutaneous fat and skin quality, respectively. The single closed square represents mean score and the error bar represents the standard deviation. The significant difference of mean score of any compared groups is represented by the asterisk.

brown color (FAS) or dark red-brown color (SSS, TS), suggesting the inappropriate soft cadaveric appearances. The well-preserved nerves and vessels obtained from all three methods were noted with more structural rigidity detected in the FAS-embalmed rabbit. Moisture contained in the TS- and SSS-embalmed rabbits reflected enough hydration which was opposite to the dehydration effect of formaldehyde seen in the FAS-embalmed rabbit (Brenner, 2014; Hunter *et al.*, 2014).

The study of muscle and tendon histology revealed muscle fiber fragmentation in the TS and SSS-embalmed, but not in the FAS-embalmed rabbit. The fragmentation was also seen in the tendon of the TS-embalmed rabbits. This result corresponded with the report of Thiel solution affected muscle fiber fragmentation in human body and this effect arose from the corrosive components such as boric acid and salts, thereby increasing joint mobility (Benkhadra *et al.*, 2011). In the present study, muscle fiber fragmentation was also demonstrated in SSS-embalmed rabbits. This phenomenon could be due to the saturated NaCl that can cause protein denaturation and precipitation. Shirai *et al.* (2015) also reported muscle disintegration in SSS-embalmed cadavers. Although the fragmentation appearance of the muscle was different between the TS- and SSS-embalmed rabbits, this could explain why the TS- and SSS-embalmed rabbits had greater joint mobility which was reflected by the ROM values. The ROM values of TS- and SSS-embalmed rabbits were in correspondence with those reported in Hayashi *et al.* (2014)

and Balta *et al.* (2019) that the TS- and SSS-embalmed cadavers have greater ROM values compared with FAS-embalmed ones. Interestingly, the hallmark evidence of tendon fragmentation was detected in TS- but not SSS-embalmed rabbits. This result is partly consistent with the study of Benkhadra *et al.* (2011), suggesting that the flexibility of Thiel embalmed cadavers is histologically correlated with muscle fiber fragmentation. It was noted that the evaluated satisfaction scores of tendon consistency of the FAS-, TS- and SSS-embalmed rabbits were not significantly different.

According to the microbial growth determination, samples collected from the pharynx, pleural cavity, abdominal cavity, and rectum were sufficient to reflect bacterial and fungal growth throughout the rabbit body as they are commonly tested sites in human cadaver (Hayashi *et al.*, 2014). The FAS showed more pronounced effect in eliminating bacterial and fungal growth than those of TS and SSS. The possible explanation is that the TS contains only propylene glycol as a disinfectant (Brenner, 2014) while those of SSS were formaldehyde and phenol. However, we speculate that the low concentration of formaldehyde in the TS- and SSS-embalmed rabbits could explain why infection was still detectable. In accordance with our results, the study by Osman, Abdeen, Edriss & Suliema. (2014) revealed the relationship between formaldehyde concentration and fungal growth in cadavers. The authors found that high formaldehyde

concentration (70%) of embalming fluid yields no infection while low concentration (10%) gains 5 fungal species infected to the cadaver, suggesting that formaldehyde concentration is one of the crucial factors combatting the microbial infections. Another possible reason for infection was the storage condition as our study was designed to keep all rabbit bodies in a transparent sealed plastic bag at ambient temperature. Many previous studies, kept the soft cadaver in cold temperature for the best storage condition (O'Sullivan & Mitchell, 1993; Sangchay, 2014); nevertheless one publication by Hayashi *et al.* (2014) reported the storage condition at room temperature. In this study, the main reason we kept the embalmed rabbits at ambient condition was due to the plan for PSU embalming fluid preparation, which will be expected to serve the learning of human gross anatomy for the second year medical students. Because a great number of cadavers were used in each year, keeping all cadavers in cold temperature is considerably difficult in practice. Therefore, the embalming fluids, TS and SSS could be selected to produce satisfactory soft cadaver at room temperature, which is a higher temperature than that of Hayashi *et al.* (2014). Additionally, the weather in Songkhla province is extreme as there were both rainy and sunny periods. The high temperature and humidity during the experiment caused bacterial and fungal infection with colonies detected in TS and SSS-embalmed rabbit bodies; however, those colonies could not be detected by the naked eye.

The limitation of our preliminary study was the small sample size that could not reveal more qualitative measurements of certain parameters. As the well-known formaldehyde embalming technique was not our major target, the single FAS rabbit then could be used only for descriptive purpose. Statistical analyses of embalming properties of all fluids would further strengthen the results of study but it requires a larger sample size. Additionally, this study was a single time point (3 months) investigation of the property of embalming fluids. Data retained from longer period or multiple time points of study may possibly provide more varied outcomes. Lastly, all participants including graduate students, scientists and researchers as well as 2<sup>nd</sup> year medical students previously experienced only the formaldehyde embalmed human cadaver. In evaluating the rabbit bodies embalmed by either TS, SSS, and FAS might be new to all participants. However, as a preliminary study, the embalming properties of all solutions were successfully revealed with insights to further assist the PSU cadaveric embalming project.

In conclusion, this study examined the properties of three embalming fluids. Our present observations were mainly focused on the TS and SSS as these formulas have been well studied and thus, could provide us the fundamentals in formulating the PSU embalming fluid.

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