

The Processing and Characterization of the New Semi-absorbable Bone Wax Made from Rice Starch Blended with Beeswax

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Abstract:

Background: In the 21st century, bone wax is one of the most prevalent biomaterials to help mechanically control bleeding from bone surfaces in almost every surgical procedure. It is bio-inert, albeit a foreign body with non-absorbability, and rarely causes complications such as granuloma and persistent serous discharge. Semi-absorbable bone wax is an encouraging alternative since it highly reduces the risk of harmful response reactions on the host, which is what we primarily aim for.

Objective: This research studied the amount of medical-grade rice added to beeswax with different amounts of addition. It is used as a semi-absorbable hemostatic agent to improve the biodegradable efficiency of beeswax.

Materials and Method: Material characteristics such as scanning electron microscopy (SEM), melting point, Fourier transform infrared spectroscopy (FTIR), and water diffusibility are used to study the effect of adding rice starch powders. Bone wax is prepared from the mixture of white beeswax with isopropyl palmitate and liquid paraffin and three different compositions of rice starch powders in aqueous suspension (30, 40, and 50 wt %).

Results: The addition of rice starch powder increases the absorbability mechanism, smoothness, and whiteness and can be easily smeared on the bone surface. We have concluded that an optimized composition of 40 wt % rice starch powder has adequate quality for utilizing it as a semi-absorbable bone wax.

Conclusion: Rice starch that is incorporated into bone wax is more bio-absorbable than the original bone wax itself and will furthermore undergo additional testing in animal laboratories.

Keywords: Beeswax, Bone wax, Rice starch, Semi-absorbable, Hemostatic agent

Introduction

Bone wax is a traditional material used by general surgeons and is used to control massive bleeding from the bone surface during surgery. Bone wax was developed by Horsley in 1892. Its ingredients comprise white beeswax of 7 proportions, almond oil of 1 proportion, and salicylic acid of 1%.¹ Bone wax is sterilized by boiling and preserved in stopper bottles. Parker is the first person to use bone wax.² Bone wax consists of white beeswax 80%, liquid paraffin 10% and isopropyl palmitate 10%, which is duplicated from commercial non-absorbable bone wax (Ethicon® Bone Wax, Johnson & Johnson Co., Ltd., US). Although bone wax is effective in controlling bone bleeding. However, the disadvantage of widely used bone wax is non-resorbable. After using bone wax to stop bleeding. Bone wax remains in that location for an indefinite period and may interfere with bone healing and bacteria clearance. This can result in bone infection. Therefore, this research has an idea to develop bone wax with resorbable properties by using fillers obtained from Thai agricultural materials, which are easy to buy and cheap, such as rice, cassava, etc., and medical grade rice starch is used for work in the pharmaceutical industry.

Starch is a polymer that can be completely degraded, and is cheap when compared to other degradable polymer. Pharmaceutical grade purified rice starch powder (Era-Tab®, Erawan Pharmaceutical Research and Laboratory, Co., Ltd, Thailand) (RS) has been used in biomaterial applications.³ Recently, modified starch has been used as a hemostatic agent due to its

water-absorbing properties and when used as a filler in composites. It prevents low molecular weight materials from absorbing blood. It may concentrate platelets and clotting proteins and thereby enhance the external blood clotting mechanism. In this research, we used bone wax as a matrix material. A composite material was prepared from our bone wax and RS with various compositions.

Materials and Methods

Materials

White beeswax (British Pharmacopoeia grade) was purchased from Bronson and, Jacobs International Co., Ltd, Thailand. Liquid paraffin was purchased from S. Tong Chemical Co. Ltd., Thailand. Isopropyl palmitate was purchased from Fluka, Chemika, Switzerland. Era-Tab® (RS) was purchased from Erawan Pharmaceutical Research and Laboratory, Co., Ltd., Thailand.

Samples preparation

Bone wax itself is composed of 80% white beeswax, 10% liquid paraffin, and 10% isopropyl palmitate was prepared through a laminar air flow hood. All composition was weighed with an analytical balance. Then, melt beeswax at 80-100°C for 5 minutes. While the beeswax is melting; blend liquid paraffin and isopropyl palmitate at 60°C for 5 minutes. RS is then added at 30, 40, and 50 wt % as shown in Figure 1. The solution is then stirred for 10 minutes until bone wax is well incorporated with RS. It is then stored and contained in a mold and cooled down to room temperature.

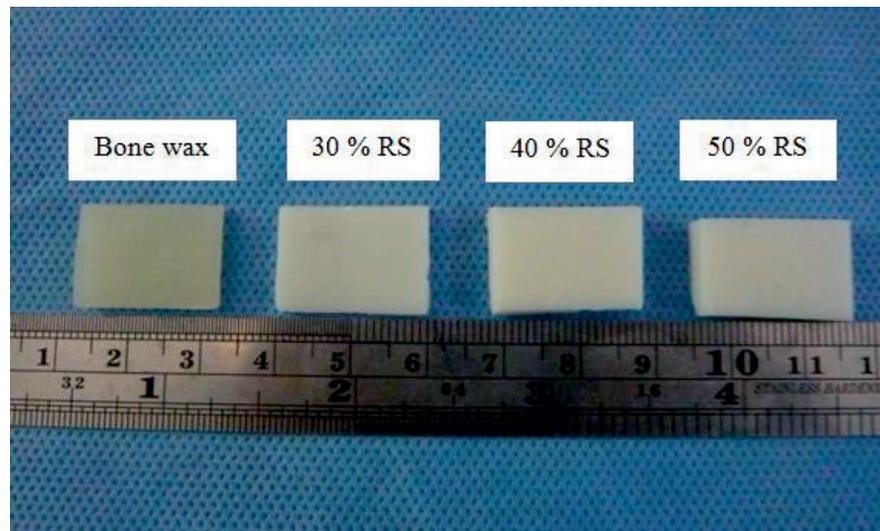


Figure 1 Digital photo of different samples

Characterization

- SEM/EDS

- SEM (JSM-6335F, JEOL, Tokyo, Japan) Energy dispersive spectroscopy (EDS) was used to analyze the morphology of Era-Tab. On the other hand, the light microscope was used to investigate the morphology of the composite surface.

- Thermal analysis

- Thermal analysis was determined by a differential scanning calorimeter (DSC7, Perkin-Elmer). The sample was heated under a nitrogen atmosphere from 20 to 120°C using a scanning rate of 5°C per minute.

- FTIR analysis

- FTIR (FTIR, Thermo Nicolet) was used to characterize the composition and functional group of materials.

- Water diffusibility

- This method was adapted from the equation.⁴ Each of the samples was accurately weighed (W1) and separately immersed in distilled water at room temperature for 24 hours. The swollen samples were removed and the excess water

was wiped off from the surface. Then, the swollen samples were reweighed (W2). The percent water diffusibility (S) of the samples was calculated using the following equations.

$$S (\%) = \frac{(W2-W1)}{W1} \times 100$$

- Statistical analysis of data was collected from three samples that showed average and standard deviation values.

Results and Discussion

This result was found that RS had an average particle size of about 5 to 6 microns (Figure 2). The morphology of the sample surface with 40 wt % of RS showed the particle distribution matrix of bone wax (Figure 3). The chemical compositions of RS powder contained 58.39 ± 1.54 wt % of carbon and 41.61 ± 1.61 wt % of oxygen (Figure 4).

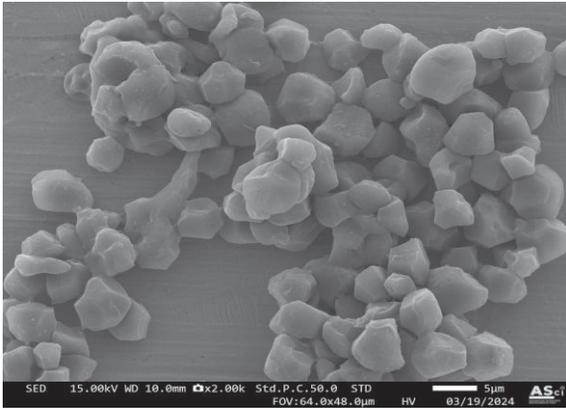


Figure 2 SEM of RS powder



Figure 3 Light micrograph of an additional 40 wt % of RS with magnification of 10X

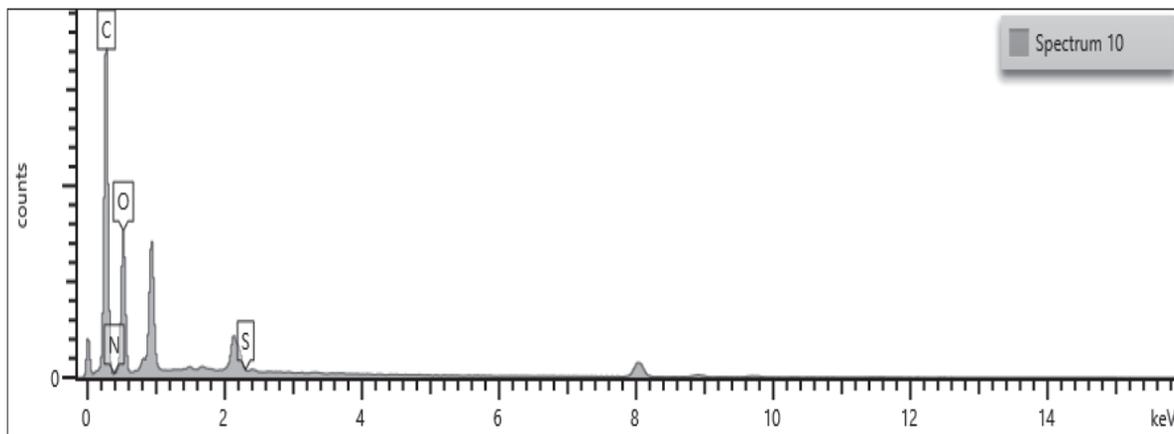


Figure 4 EDS of RS powder

Figure 5 shows the melting point of bone wax increased from $61.52 \pm 0.05^\circ\text{C}$ to 63.72 ± 0.04 , 64.38 ± 0.05 and $65.52 \pm 0.06^\circ\text{C}$ when content of RS increased was 30, 40

and 50 wt %, respectively. This was possibly due to the prevention of starch granules from thermal treatment.

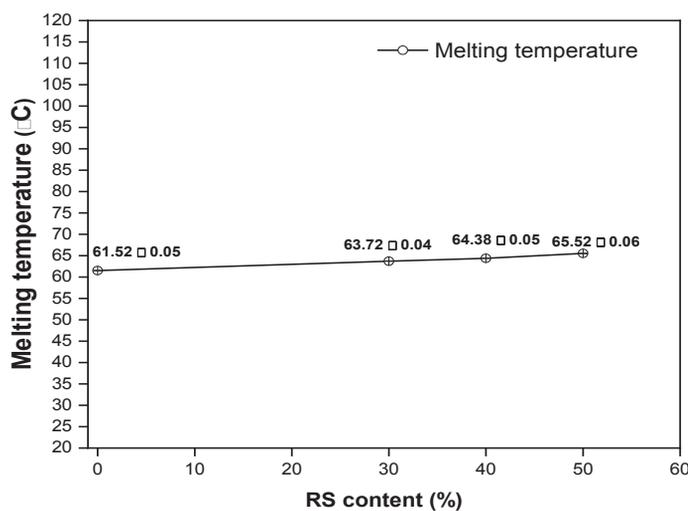


Figure 5 Effect of RS content on the melting temperature of the samples

The FTIR spectra of bone wax, RS, composites are shown in Figure 6, and the most important vibration modes were the C-H stretching around 3000 cm⁻¹ and the -CH deformation modes around 1460 and 1380 cm⁻¹. The atoms directly attached to the aliphatic groups may result in significant shifts from the standard frequencies. In particular adjacent atoms with high electronegativity will shift the band locations to higher frequencies. When two methyl groups were on a single carbon (isopropyl) band of approximately equal intensity, it occurred at near 1390 and 1365 cm⁻¹. The presence of the t-butyl group can be

confirmed by the presence of bands around 1255 and 1210 cm⁻¹ while the isopropyl group showed bands near 1170 and 1145 cm⁻¹. When there were four or more CH₂ in a row, a rocking absorption was found centered at 720 cm⁻¹. The FTIR spectra of RS showed the prominent peaks at 3300, 2933, 1643, 1156, 1019, 928, and 861 cm⁻¹ which were -OH stretching (3593 to 3643 cm⁻¹), -C-O stretching, and -OH deformation vibration (1050 to 1200 cm⁻¹). The FTIR spectra of the addition RS from 30 to 50 wt % showed that no interaction between bone wax and RS during the physical mixture.

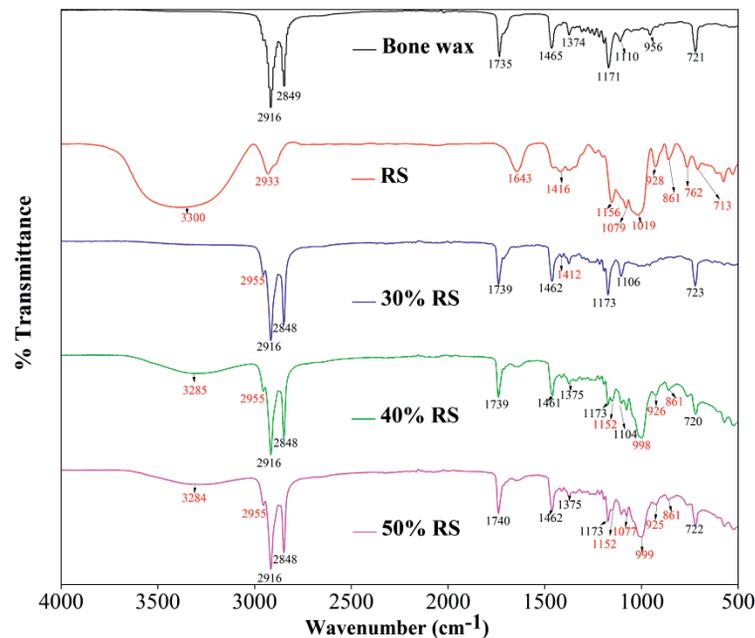


Figure 6 FTIR of bone wax, RS, and different compositions

Figure 7 shows that the water diffusibility of bone wax was constant with an increase in dwelling times due to its non-absorbability. However, RS absorbs water into its structure and swells, thus the addition of RS in the bone wax increased the percentage of diffused

water of 30 wt % RS as 0.2 ± 0.22 %, 40 wt % RS as 0.67 ± 0.33 % and 50 wt % RS as 1.47 ± 0.21 % for 80 h. This result showed that 50 wt % of RS in bone wax had the highest percentage of water diffusibility.

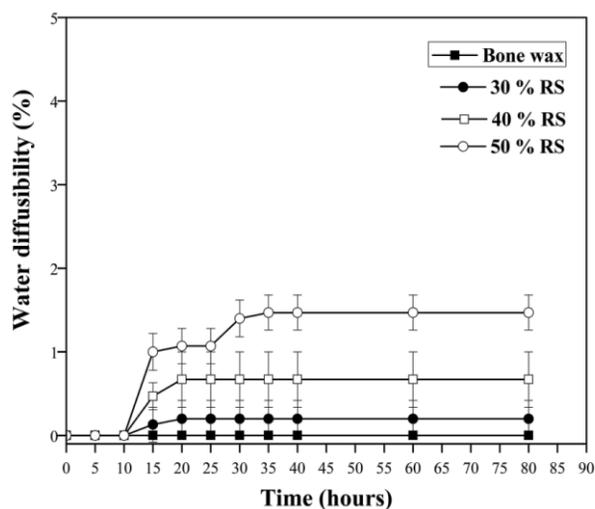


Figure 7 Effect of RS starch content on water diffusibility vs various times

Increasing the content of RS in bone wax increased the melting temperature of the samples due to the obstruction of RS in the bone wax structure. This was possibly due to Era-Tab particles being highly stable and tend to agglomerate. It was used to increase the amount of tablets. There is a wide endothermic temperature range from 70 to 120° C, which may make the melting temperature range of this experiment inconsistent with other studies. The sample that contained RS 40 wt % provides pliability by hand which is similar to commercial bone wax and has potential use as a novel semi-absorbable bone hemostatic wax. However, the effects of water diffusibility of 40 wt % RS are lower than 50 wt % RS due to the amount of RS that is still high on the surface of the bone wax. Water diffusibility of 40 wt % RS has a low percentage because most of the RS content is embedded in a matrix of bone wax. However, commercial bone wax is a bio-inert in the human body. Several studies reported the influence of bone wax on osseous defects. The combination of inhibited osteoblast activities and physical barriers may prevent bone healing and an increased risk of infection.⁵ On the other hand,

Rockwood, et al⁶ implanted bone wax in the bones and muscles of experimental rabbits and they found that the tissue reaction included; 1. No infection 2. Minor absorbability was seen after six months of surgery and inhibition of bone healing, and 3. Minor inflamed reaction on tissue, compared to suture.

Conclusion

A novel semi-absorbable bone wax is softness and pliability to stop bleeding from the bone. The optimized composition was the addition of 40 wt % RS powder into bone wax. Hence the fact that there is more bio-absorbability than the original bone wax and this research should be further tested in animal laboratories.

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