

NONWOVEN GREIGE COTTON FOR WOUND HEALING AND HYGIENIC PRODUCT APPLICATIONS**J. Vincent Edwards****Nicolette Prevost****Elena Grave****Brian Condon****USDA-ARS-Southern Regional Research Center****New Orleans, LA****Lawson Gary****Edmund Carus****T.J. Beall Company****Drew, MS****Abstract**

The potential to use greige (non-bleached) cotton in nonwoven absorbent products has received increased attention. This is due to innovations in cotton cleaning and nonwoven hydroentanglement processes that open and expose the hydrophilic cellulosic component of greige cotton fiber to water absorption while retaining the cuticle waxes and pectin. Wound healing and hygienic value in greige cotton has been recently demonstrated and will be discussed in this presentation. Greige cotton demonstrated accelerated clotting over bleached cotton, and generates low-level hydrogen peroxide which is associated with accelerated formation of granulation tissue in wound healing. In addition mechanically cleaned surfactant –treated nonwoven greige cotton functions similarly or better than commercial diaper coverstock layers. Tests measuring rewet, strikethrough, and acquisition of synthetic urine, and comparing commercial samples with industry standards of pre-clinical diaper performance, revealed that nonwoven greige cotton can function as a top sheet in diapers and other incontinence products.

Introduction**Cotton in Nonwovens**

In recent years the preference to use cotton fibers in nonwoven absorbent products has increased. Cotton fiber is naturally renewable and biodegradable. Cotton's characteristic soft hand, hypoallergenic properties, absorbency, and cellulosic composition have been historically utilized mostly in woven fabric products. Cotton's current use in nonwovens is estimated to only be approximately 2% (by volume/weight) of the total fiber consumption in nonwovens. However a recent study by Lutel et al. puts the nonwoven consumption of cotton at a higher level (Lutel et al., 2013). Most of the cotton used at present in absorbent nonwovens is bleached cotton including lint, gin motes, linters, comber noils and so-called other cotton textile processing wastes. However, the potential to use greige (non-bleached) cotton in nonwoven absorbent products has received increased attention based on innovations in cotton cleaning and nonwovens processes that open and expose the hydrophilic cellulosic component of greige cotton fiber to water absorption (Sawhney et al. 2010, Condon et al, Sawhney et al. 2011). Here we show how it possesses structure/function properties suitable for advanced wound healing and hygienic applications.

The Cotton Fiber

The morphology of the cotton fiber (Figure 1) consists of a cuticle which contains lipids (hence hydrophobic) that are only approximately one percent by weight of the fiber (Wakelyn, 2007). The primary cell wall below the cuticle contains pectin, cellulose and some proteinaceous material, and the secondary cell wall is where most of the cellulosic content of the fiber resides. Cotton cellulose is characterized as a microfibrillar structure that contains cellulose crystallites and at the most fundamental level cellulose chains which are packed closely through hydrogen bonding.

The Cotton Fiber

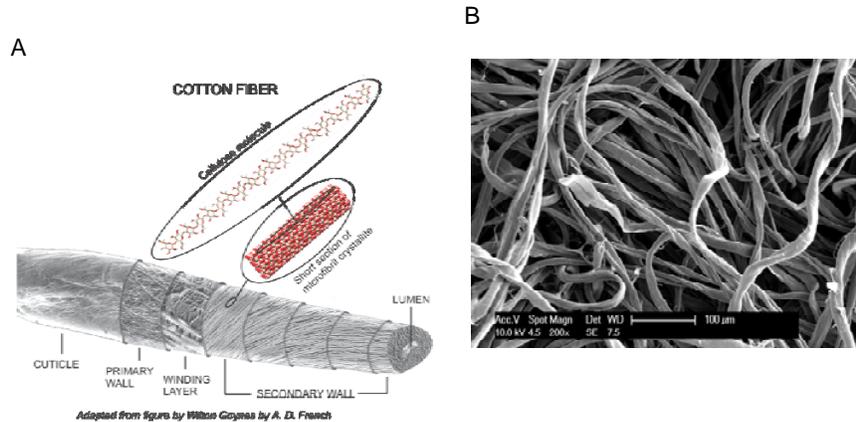


Figure 1: (A) Montage of cotton fiber assembled as scanning electron microscopic images of distinct layers of the fiber. (B) SEM of hydroentangled greige cotton (fiber image montage is a rendering courtesy of Wilton Goynes and as modified by Dr Alfred French).

During processed bleaching and scouring of cotton the cuticle and primary cell wall are removed leaving primarily the cellulosic secondary cell wall of the fiber. This form of scoured and bleached cotton has been used solely in dressings for over a hundred and fifty years. However, in recent years it has been shown that mechanically cleaned nonwoven greige cotton, which still retains the cuticle and primary cell wall is suitable for use in absorbent nonwoven materials alone or in combination with hydrophilic fibers. Pectin is present in greige cotton in low percentage, by-weight amounts 0.6 – 1.0%, and has properties that support accelerated granulation tissue formation.

The following outlines some highlights contrasting greige cotton with scoured and bleached cotton.

Aspects of Greige Cotton for Wound Healing and Hygienic Uses

- Greige cotton cleaned to a high standard of purity and hydroentangled to make absorbent nonwovens.
- Lower processing cost.
- Lower carbon footprint than bleached cotton due to absence of finishing processes.
- R&D investigations on the advantages of pectin and waxes in greige cotton in wound healing (8).
- Processes very well in textile nonwoven procedures.
- Transmits synthetic urine more rapidly than scoured and bleached cotton (Edwards et al. 2013).
- Have good rewet and strikethrough properties.

Aspects of Bleached and Scoured Cotton for Wound Healing and Hygienic Uses

- Lower carbon footprint than synthetics.
- Native waxes and pectin are absent or mostly stripped away.
- Used for over 150 years in medical gauze dressings.
- R&D work has achieved an FDA-approved treated dressing for chronic wounds (Edwards et al).
- Costs more than greige cotton to process.
- Historically pursued for its high cellulosic content as an absorbent.
- Does not tend to have good rewet properties.

This report addresses the analysis of the role of mechanically cleaned nonwoven cotton as a new material that can be used in dressings to modulate wound healing. It is thereby envisioned, as this report suggests, that greige cotton has the potential to enhance treatment modalities for non-healing wounds where the stages of hemostasis, inflammation, and stalled cell proliferation are an issue.

Here we address that cotton's pectin and wax modulate functional properties giving; 1) increased rate of clot formation 2) low level hydrogen peroxide.

Materials and Methods

A commercially available bale of pre-cleaned greige cotton (True Cotton™) was acquired from T. J. Beall, LLC. A test matrix consisting of various types of hydroentangled nonwovens containing mechanically cleaned greige cotton fiber were produced in trials with Trützschler Nonwovens at their pilot facility in Wolfsgartenstraße 6, 63329 Egelsbach, Germany.

Wound Healing Materials Experiments

Hydrogen peroxide production from nonwoven greige cotton was assessed with scopoletin assays to determine the efficacy to generate low-levels conducive to wound healing (Schmidt et al., 1993). Pectin levels in the cotton samples were determined through hydrolysis to the constituent galacturonic acid monomers and quantitative assessment with colorimetry. Greige cotton samples (50mg in 5mL) were subject to enzymatic hydrolysis for 48 hours in a 0.5% viscozyme (Sigma Chemical mixture of hydrolases) in sodium acetate pH 4.8. One hundred microliters of sample and 100 uL of saturated copper sulfate were mixed and place in a dry bath at 100C for 30 minutes whereupon 0.8 mL of diluted Folin-Ciocalteu reagent was added. An immediate color change signaled the presence of galacturonic acid which was quantitatively assessed.

Thromboelastography (TEG) was employed to assess clotting activity of materials of this study under the viscoelastic properties of whole blood with low shear conditions. TEG provides information about global hemostatic function from the beginning of clot formation through clot retraction and fibrinolysis. Below the relationship between a qualitative tracing and the quantitative parameters including R, K, and MA are shown. TEG identifies the relative contribution of clotting factors and platelets to the overall coagulation process. TEG is being increasingly used for the evaluation of biomaterials on blood coagulation (Peng, 2010) and for the purposes of this study was employed to compare the difference in clotting performance of bleached and scoured cotton versus greige cotton as well as greige cotton as a carrier of hemorrhage control agents as silicon dioxide.

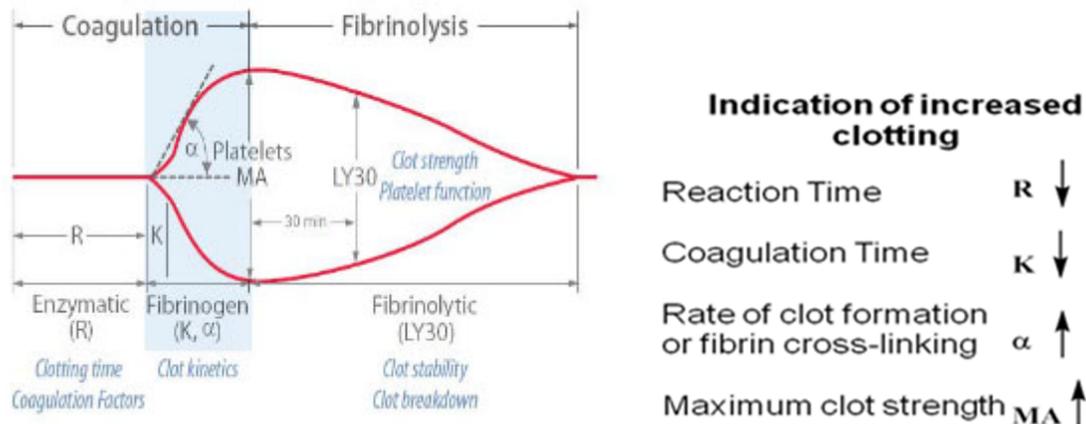


Figure 2: Thromboelastography profile of clotting events.

Hygienic Incontinence Material Performance Experiments

To obtain penetrant-treated samples mechanically cleaned nonwoven greige cotton was treated by Huntsman Chemical with penetrant (Ultra Fill CO) to 53 – 87% wet pick-up, and then dried at 140°C for one minute. The final add-on was 0.8% solids. The penetrant (a polyurethane/polysiloxane formulation) used was employed to facilitate more rapid strike through times and acts to promote hydrophilic transport of the liquid to the absorbent core.

Marketing Testing Strategies, Kalamazoo Michigan performed liquid strike-through, rewet, and liquid acquisition tests. The Liquid strike-through Time Test is an EDANA recommended test method. Strike through Time is measured according to EDANA/INDA WSP 70.3 (former ERT 150) and WSP 70.7(former ERT 153). This test electronically measures the liquid-strike-through time. The liquid-strike-through time is the time taken for a known volume of test liquid (simulated urine) applied to the surface of a test piece of nonwoven coverstock, which is in contact with underlying standard absorbent pads to pass through the nonwovens. Rewet properties were assessed according to EDANA/INDA WSP 80.10 (former ERT 151) and WSP 70.8 (former ERT 154); this test electronically measures the wet back of synthetic urine through a prepared sample onto a filter paper. After applying a defined amount of liquid on the prepared sample (strike-through-time test), a simulated baby weight is automatically lowered onto the specimen with a speed defined by EDANA/INDA, and remains there for a specified period of time. Through a special filter paper and an electronic balance, the amount of liquid is determined, which - due to the load - is passed back through the specimen's surface into the filter paper. The acquisition test evaluates in real time how well liquids get into the absorbent core at any flow rate: 0.9% saline solution is applied to the sample at a specific volume and flow rate. In the test results, time in seconds from the start of the test is shown on the X-axis, while the Y-axis shows the total number of ml of both leakage (or fluid acquisition) and overflow.

Results and Discussion

Here we outline results on the wound healing and hygienic properties of greige cotton nonwovens.

Wound Healing Properties of Greige Cotton Nonwoven

The wound healing properties of greige cotton are described here in terms of hemostatic and hemorrhage control design that addresses the rapid acceleration of clotting in an acute wound during the initial seconds of trauma, and low level hydrogen peroxide generation which is essential to wound healing events from the hemostatic through the inflammatory stage of wound healing. Previously we have shown that mechanically cleaned, greige cotton (unbleached) is a useful material for initiating hemostasis and low level hydrogen peroxide generation (Edwards et al., Wound Repair and Regeneration, 2013, 2 (12), A21). In this paper we show clotting profile results on greige cotton versus a commercial product for hemorrhage control used by the Armed Forces. Silicon dioxide promotes rapid clotting of uncontrolled hemorrhages. Greige cotton was examined as a carrier of silicon dioxide, and previously has been compared with Quick Clot Combat Gauze (CG) (Edwards 2013).

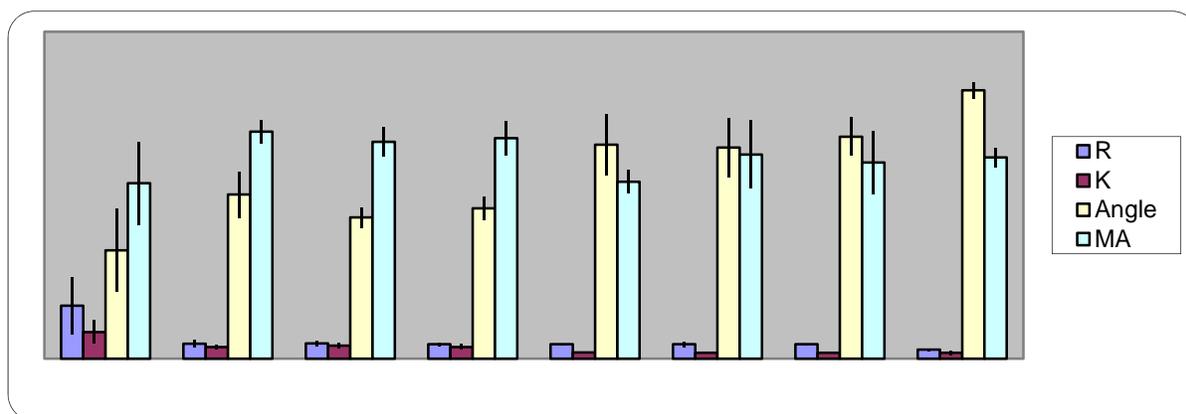


Figure 3: Thromboelastography results for silicon dioxide impregnated greige cotton; 1- blood with citrated saline (control); 2 greige cotton nonwoven (UC); 3- 60% UC, 40% N; 4- 40% UC, 60%; 5 - UC, SiO₂; 6- 60% UC, 40% N, SiO₂; 7- 40% UC, 60% N, SiO₂; 8- SiO₂, alumina doped. UC= greige cotton; N = Nylon.

As seen in Figure 3 hydroentangled greige cotton impregnated with silicon dioxide (97 percent add-on) gave similar results in the thromboelastography profile to that found with silicon dioxide alone. Sample 5 (UC) shown in Figure 3, which is one hundred percent greige cotton, also accelerated clotting two-fold more than bleached cotton. Although greige cotton nylon blends were also good carriers there was no marked improvement over greige cotton.

Pectin-Based Low Level Hydrogen Peroxide Generation

Pectin is found in a variety of semi-occlusive dressings. The role of pectin in low-level hydrogen peroxide generation has been previously documented in semi-occlusive dressings (Schmidt, et al. 1993), and has been the subject of renewed interest in dressing development. Pectin is a major structural cell wall polysaccharide found in plants (Caffall, 2009). It has been studied and used in a variety of biomedical applications in recent years for delivery of therapeutic drugs and genes, in tissue engineering, and wound healing (Munarin, 2012). Moist wound healing accelerates wound healing over a desiccated wound (Winter, 1962). In the context of wound healing the molecular features of pectin that most influence gelling, and hence moist healing are molecular weight, and both degree and pattern of esterification. The so-called ‘egg-box’ motif that arises from calcium chelation by galacturonic acid carboxyls found in pectin is also a feature that through the mechanism of electrolyte exchange in wounds promotes non-adherence as is the case with alginate. Commercial chronic wound dressings were contrasted with greige cotton wound dressings for their relative pectin-dependent hydrogen peroxide generation levels. As shown in Figure 4 pectin levels in commercial dressings including Duoderm patch and paste were found to be ten times the levels observed in greige cotton (0.59-0.53 mg/gram versus 0.04 – 0.060 mg/gram respectively).

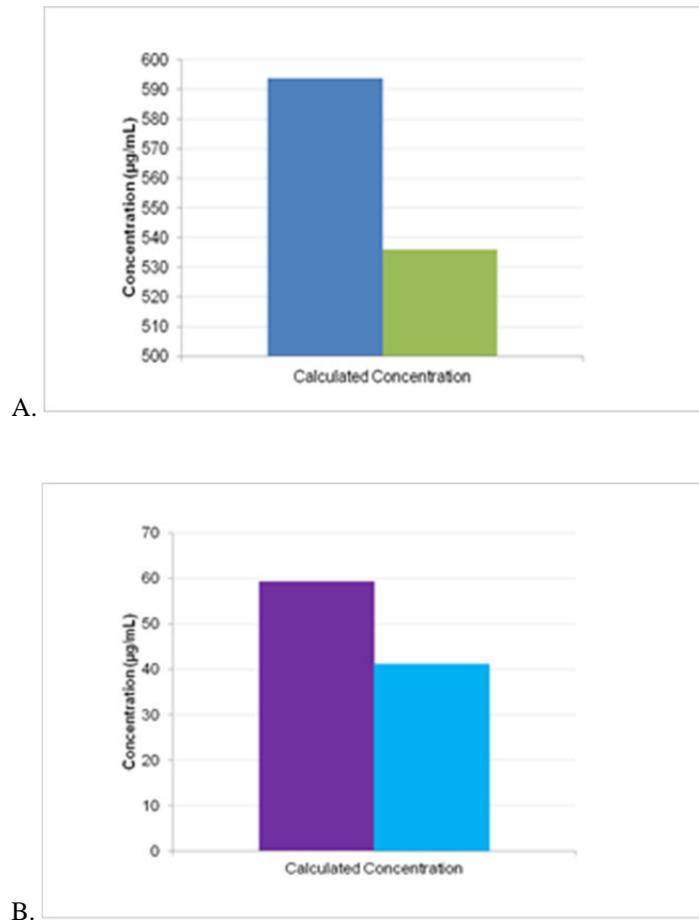


Figure 4: Pectin levels in Duoderm patch (blue) and Duoderm paste (green) (A) and in needle punched (purple) and hydroentangled nonwoven cotton (light blue) (B). Concentrations of hydrolyzed pectin are shown for galacturonic acid as detailed in the Materials and Methods section. Note axis ranges are not scaled between the figures.

Low-level Hydrogen Peroxide Generation from Greige Cotton

In recent years the role of low level hydrogen peroxide generation in stimulating wound healing as an important signaling event in cell proliferation in every stage of wound healing has been made abundantly clear through numerous reports on its role in hemostasis, facilitating platelet aggregation, stimulating platelet derived growth factor, and its role in inflammation inducing neutrophil and macrophage chemotaxis, and the role of vascularization with induction of vascular endothelial growth factor (Chandram, 2003; Loo, 2012; Roy, 2006).

As shown above the pectin levels associated with hydrogen peroxide generation in the commercial hydrocolloid samples had roughly 10-fold more pectin on a per weight basis. However, the hydrogen peroxide generation of unbleached cotton was found to exceed that of the commercial products on a per weight basis by two-fold. The hydrogen peroxide levels were found to vary as a function of the intact primary cell of the fiber. Thus the mechanism of plant fiber hydrogen peroxide generation appears more robust than commercial pectin within the frame work of low-level criteria for wound healing.

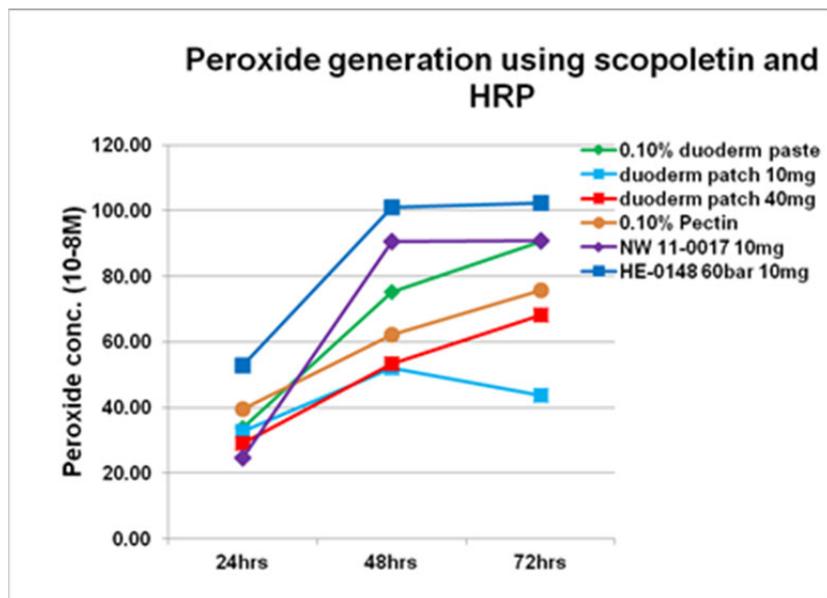
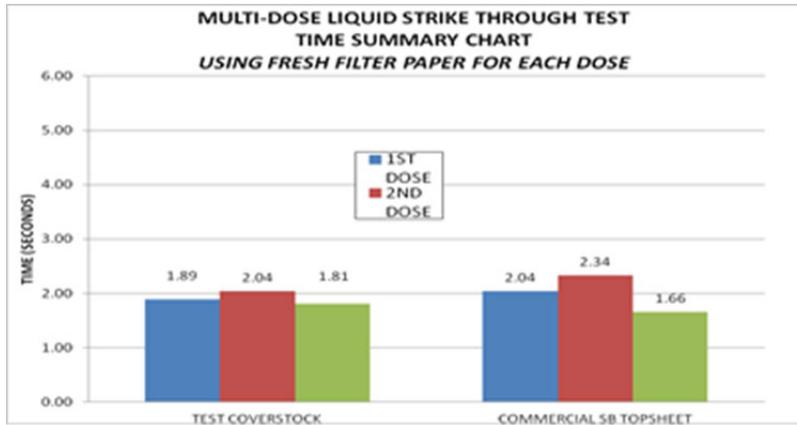


Figure 5: A plot of hydrogen peroxide generation over a period of three days from different commercial dressing material, pectin, and hydroentangled and needle punched greige cotton. NW = needle punched cotton; HE = hydroentangled.

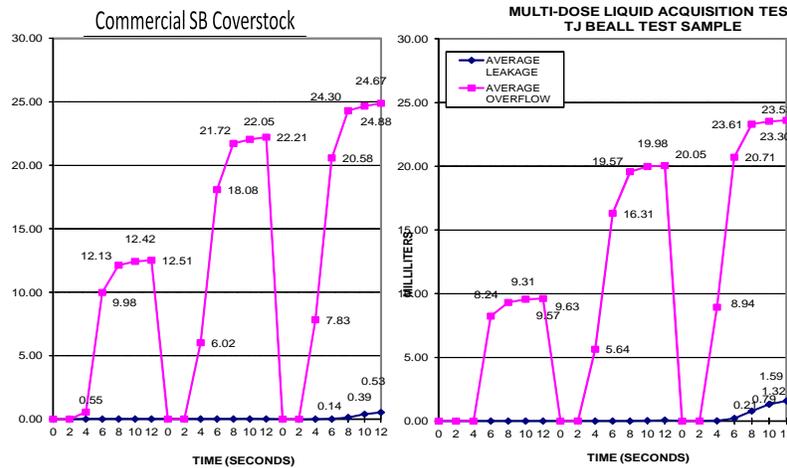
Studies on Penetrant-Treated Mechanically Cleaned Greige Cotton

The performance of penetrant-treated mechanically cleaned greige cotton in the multiple strike-through tests as shown below (Figure 6) was similar to commercial samples when fresh filter paper was used. However, considerable improvement was observed over commercial samples when the same filter paper was used. Rewet increased some over untreated samples with addition of penetrant to the cotton, but it was still half the volume of the commercial sample used as a comparison in the test. One of the most remarkable improvements from the penetrant-treated samples was the improvement in fluid acquisition (see side-by-side comparison in Figure 6) over untreated samples shown previously. As seen below the acquisition overflow and leakage were significantly less than commercial samples following multiple insults. Thus, this series of performance tests of rewet, strikethrough and acquisition with penetrant-treated greige demonstrate the greige cotton nonwoven fabrics to be highly functional when compared with commercial spun bonded polypropylene samples.



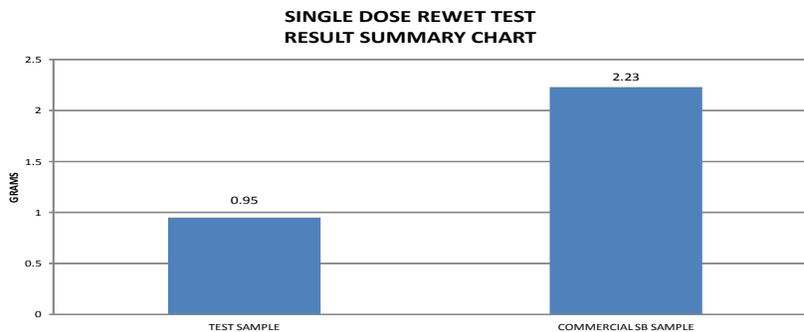
A.

Acquisition Data of Greige Cotton Compared to Commercial



B.

Penetrant-Treated 100% Greige Cotton



C.

Figure 6; Greige cotton versus commercial sample results; A) Results of strikethrough analysis left side greige cotton & right side commercial spunbond sample ; B) Liquid acquisition (x-axis, minutes & y-axis is volume, mL); left side commercial spunbond sample and right side greige cotton nonwoven; C) Rewet (y-axis grams) x-axis – l-side greige cotton, r-side commercial spunbond control.

Summary

Greige cotton has excellent procoagulant properties and promotes clotting of whole blood at a two-fold greater rate than bleached cotton. Greige cotton as a hemostat and carrier acts synergistically to enhance silicon dioxide mediated clotting. It demonstrates thromboelastograph clotting profiles consistent with potential hemorrhage control properties that are better than or comparable to Quick Clot Combat Gauze. Greige cotton hydroentangled samples performed similarly or slightly better in generating hydrogen peroxide levels previously implicated in stimulating healing as found in the semi-occlusive dressing DuoDerm. Generation of low-level hydrogen peroxide by hydroentangled nonwoven greige cotton is consistent with the retention of accessible levels of pectin correlated to enhance granulation tissue or improved cell proliferation and fibroblast and macrophage production.

Results suggest that highly cleaned greige (but not scoured and bleached) cotton nonwovens possess properties that may improve incontinence product maintenance of dryness and promote skin care in absorbent products. Results have shown that mechanically cleaned greige cotton can now be considered as a low-cost bio-based fiber alternative to synthetic fibers for top sheet use and design. The results of our study show that mechanically cleaned cotton represents a bio-based, eco-friendly alternative to synthetic fibers when designed into the layers of incontinence products. Mechanically cleaned greige cotton retains the cotton fiber cuticle lipids (hence it is more hydrophobic than scoured and bleached cotton) and primary cell wall pectin, which are removed in scoured and bleached cotton. Although bleached and scoured cotton afford highly absorbent properties for many applications it does not typically have good rewet and strikethrough properties. On the other hand we have found that mechanically cleaned nonwoven greige cotton has superior rewet and strikethrough properties. Greige nonwoven cotton also processes very well in nonwoven procedures. Additionally we have found that it transmits synthetic urine more rapidly than scoured and bleached cotton.

Acknowledgements

The authors wish to acknowledge the contribution of True Cotton™ from TJ Beall, LLC, which were used in the greige cotton work reported here; Jim Hanson and Mark Boleyn of MTS in Kalamazoo MI for running the diaper performance tests; Pat Eberlein and Nolan Starnes Huntsman Chemical for penetrant-treatment of mechanically cleaned greige cotton nonwovens used in this study.

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