

CELLULOSIC FIBER FOR ODOR AND PH CONTROL

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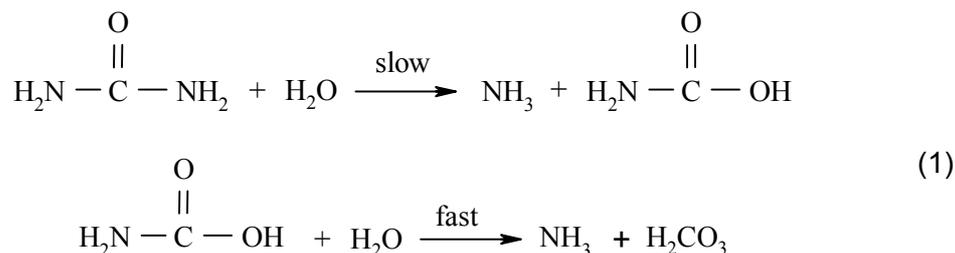
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Abstract

A new approach to odor and pH control in medical and hygiene care applications relies on a treatment of absorbent fluff with selected enzyme inhibitors. Cellulose fiber was found to be a convenient carrier for some chemicals which modify the structure of an enzyme and/or block its active site. In an absorbent system exposed for example to body liquids containing urea, selected substances, when released from the fibrous material, can effectively slow down enzymatic hydrolysis of NH_2CONH_2 and suppress the emission of ammonia. An additional benefit of this technology is in this case a control of the pH of the skin environment and keeping it closer to neutral or slightly acid region. A slowdown of the ammonia emission can be enhanced by blending sodium polyacrylate-based superabsorbent polymer (SAP) particles with the modified cellulose fibers. Suitable inhibitors of urease can also be applied directly to absorbent nonwoven fabrics comprising fluff.

INTRODUCTION

Ammonia is released from body fluids as a result of fermentation of urea, which is catalyzed by an enzyme called urease. A molecule of urea can produce two molecules of ammonia according the following reactions:



Without the presence of urease the rate of the hydrolysis of urea is relatively slow. However, the enzyme can speed up this reaction by a factor of 10^{14} . Ammonia emitted from some biological fluids such as urine, sweat and blood produces unpleasant odor and changes the pH from slightly acidic or neutral to alkaline. Higher pH is undesirable because of its adverse effect on skin, causing irritation and microbial infections.

The issue of ammonia formation is particularly serious in medical and personal hygiene areas. So far no fully satisfactory remedy has been found to address it. This is why manufacturers of adult incontinence (AI) articles, baby diapers and feminine sanitary napkins consider odor control still as an unmet need. Numerous attempts have been made by various researchers to absorb ammonia and other malodorous substances with commonly known materials such as zeolites, clay, cyclodextrin, baking soda etc. [1-3]. In general, these concepts were based on blending odor scavengers with absorbent cores and thus containing them in personal care devices. High cost and low effectiveness have been usually main drawbacks associated with such technologies. Another, rather unsuccessful approach, involved the use of fragrances to mask the odor with some aromatic formulations [4]. This solution may be effective in general only until the waste liquid matter begins to decompose. Unfortunately, pleasant smell combined with the resultant offensive odor fails to prevail as a perfume scent.

Urease is produced by various microorganisms, such as some strains of *Escherichia coli* [5], so removing these microbes from the body environment became one of the indirect ways of combating the odor of ammonia. An example of such approach is the use of ethylenediaminetetraacetic acid (EDTA) sodium salt [6], a strong chelating agent, for treating absorbent materials destined for personal

hygiene articles. EDTA can retard bacterial growth by depleting the microbes of calcium necessary for their diet and thus by limiting the amount of secreted enzymes. One needs to be cautious with applying strong antimicrobial agents in order to avoid possible adverse side effects on user's body. On the other hand, milder bactericides may not be effective enough to eliminate the target enzymes to an appreciable extent. Certain SAP manufacturers make special grades of superabsorbents with antimicrobial and/or odor-control claims. Some of these materials contain agents affecting bacterial growth [7] with potential issues mentioned above and others are based on the ability of binding ammonia by free carboxyl groups of partially neutralized cross-linked polyacrylic acid [8]. From the standpoint of maximum absorbency capacity, the optimum degree of neutralization of the polymer carboxyl groups at which the SAP particle swell to its utmost is about 70%. However, in order to enhance its ammonia-binding power the amount of free carboxyl groups needs to be higher. This leads to increased overall material cost and lowered absorbency, the primary function of the superabsorbent polymer.

The approach described in this paper is different from the prior art. The proposed concept involves incorporation of selected chemical agents to the components of disposable absorbent systems with intent to deactivate the enzymes responsible for the decomposition of body fluids [9].

MATERIALS AND METHODS

Southern softwood kraft cellulosic fibers, Foley Fluffs® (Buckeye), were treated with aqueous dispersions of various urease inhibitors at various add-on levels and used for further experiments. Similarly, some of these dispersions served for treating airlaid nonwoven fabrics composed of cellulose fibers, SAP and binding components. Descriptions of these procedures and chemical treatments are contained in the patent application [9].

The liquid chosen for the experiments described in this paper was a blend of human urine specimens donated by several healthy adults. In order to create conditions favoring ammonia production the liquid was enriched with small amount of urease (U1875, Sigma).

Ammonia emission tests were performed in specially-designed, hermetic containers maintained at constant temperature of 37°C (Fig. 1). Each container could be opened for placing in it a sample of the studied material. It was also equipped with two outlets, one for introducing the test liquid and the other for inserting the ammonia Draeger test tube (Fig. 2). After insulating the test sample with an aliquot of urine, the container would be sealed off and the experiment would be carried out for a period of time. Measurements of the concentration of generated ammonia were taken during the test at various time intervals for monitoring the kinetics of the decomposition of urea. Thus obtained results could be presented in a graphical form by plotting the amount of ammonia released to the atmosphere inside the containers against the time of the experiment. Examples of such graphs will be shown later in this text. In some experiments we also measured the change of the pH of the sample surface during the time of the test.

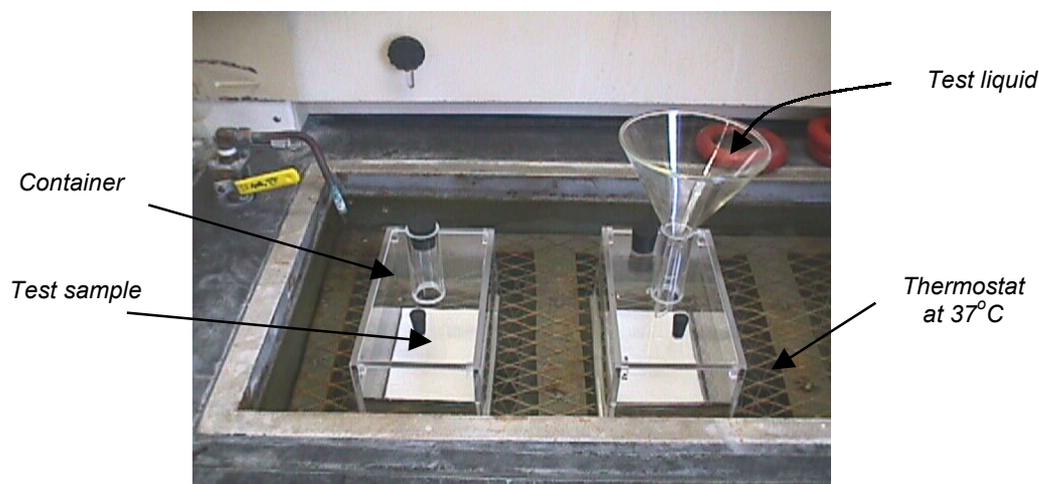


Figure 1. Ammonia emission test set up

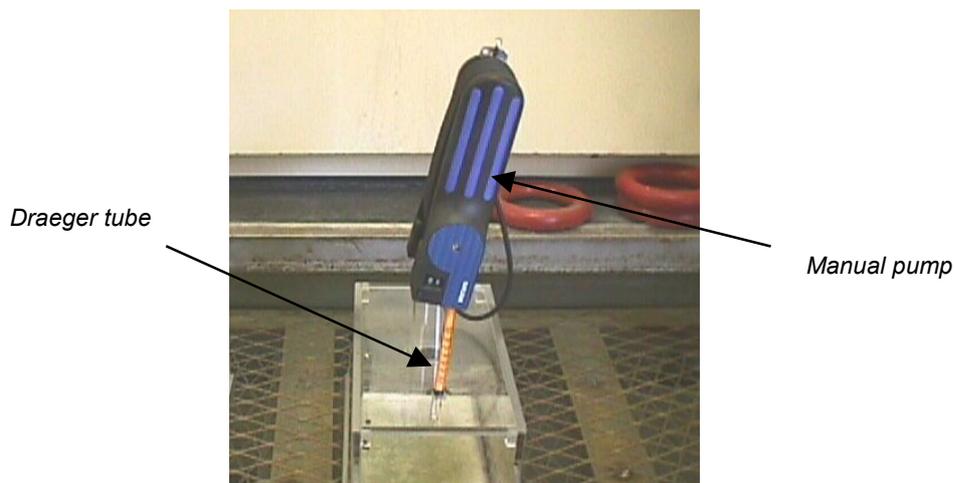


Figure 2. Manual instrument for measuring ammonia concentration in air

RESULTS AND DISCUSSION

There are various chemical compounds which can react with urease by blocking its active site or by modifying the macromolecule making it less effective for the decomposition of urea. The active site of the enzyme is shown in Fig. 3. The key elements here are two nickel atoms combined with few organic segments and associated with four imidazole rings and a molecule of water [10]. This arrangement is compatible with the structure of a urea molecule which once trapped in it becomes quickly hydrolyzed producing gaseous ammonia. In the competitive inhibition process a potent inhibitor can react with the enzyme faster than urea itself and form a stable complex with the active site or deactivate it for instance by changing its chemical composition.

A number of examples of various inhibitors described in the literature have been listed in various references, e.g. [11-14]. Some of these agents have been used in medicine for treatment of urinary track diseases [11,12], others have been tried for agricultural applications to prevent the loss of nitrogen from the soil [13]. Other potential benefits of urease inhibitors considered by several researchers are those in the animal farming area [14] for alleviating malodors resulting from the fermentation of urea and for preserving nitrogen in the biological waste for further plant fertilization.

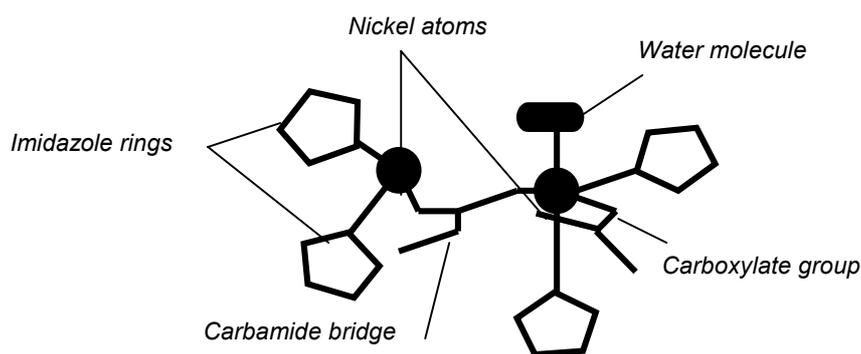


Figure 3. Active site of urease

Urine of healthy people is usually quite sterile and relatively stable. It begins to decompose more readily only after contamination with microbes producing appropriate enzymes. In theory, if no urease-producing bacteria were available in the test system we should not observe any effect of the inhibitor-treated cellulose fiber on the kinetics of ammonia emission. Such bacteria are generally ubiquitous in close-to-body environment. In our study, instead of employing urease-secreting microorganisms we added the ultimate enzyme directly to the test liquid. In order to specify practical conditions for the ammonia emission we determined at first the optimum concentration of urease in the

test fluid. The objective was to make it possible to complete the experiments within a reasonable period of time and generate ammonia in quantities easily measurable with the Draeger tube instrument.

Vizorb® 3905, a nonwoven absorbent fabric, was used as a substrate for the experiment. The test was conducted as outlined above in the previous section. The results are shown in Fig. 4. They determined the optimum concentration of the U1875 urease at 0.1 mL/100 mL liquid. If we assume these test conditions and excess supply of urea, the maximum quantity of ammonia generated in one hour due to 0.1 mL urease would be almost 70 mg, according to the activity of the enzyme (650 U/mL) specified by the vendor. Human urine contains roughly 2% urea, so in 100 mL of the liquid (maximum amount used in our tests) only 1.1 g ammonia would be obtained if, in theory, all the amount of NH_2CONH_2 were hydrolyzed. Hence, the concentration of urease selected for the study was more than enough to decompose all available urea in one hour under reaction conditions which would be ideal for the enzyme.

Every time a new urine specimen was used for each individual series of tests described below, so the actual composition of the fluid, even though taken from the same donors, could vary to some extent and therefore affect the experimental data. In consequence, the findings should be compared within each series rather than between different series of the tests.

In our experiments we could smell ammonia only when its measured concentration in the sample containers was higher than 100 – 150ppm. This was confirmed by several independent panelists of different ages and sexes. At the same time one should realize that individual test parameters may define various detection thresholds for NH_3 . To get an idea what such threshold meant in our studies, a simple calculation tells that it amounted to just about 0.3 to 0.45 μg ammonia in the test container (20cmx12cmx10cm) having a capacity of about 3 mg air inside it. So, our experimental conditions were sufficiently drastic to generate enough ammonia within several hours to be easily detected by human sense. Besides, the procedure allowed for measuring any significant reduction of ammonia due to a given the inhibitory system under investigation.

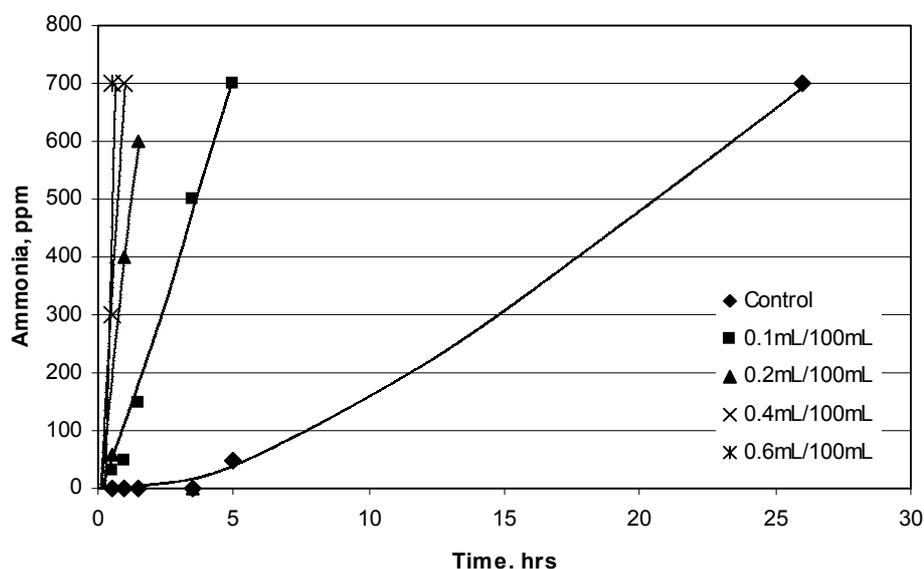


Figure 4. Ammonia emission at different concentrations of urease U per 100 mL urine from healthy male donors. Vizorb® 3905 was insulted with 100 mL liquid

Let us evaluate also any theoretical effect of the superabsorbent polymer comprised in the cellulose fiber-based composite. If the latter contained for example 15-30% SAP neutralized at about 70%, the polyacrylate salt polymer would be capable of binding at maximum 0.1 to 0.2 g ammonia though its free carboxyl groups. It is roughly 10 to 20% of the possible quantity of ammonia generated from the urea contained in 100 mL urine, provided 100% urea becomes fully fermented. The conclusion is that potentially 100% SAP material would be required in theory to neutralize much of the generated ammonia. However, it is obvious that such a solution would not be practical due to total lack of integrity of a powder-like absorbent structure.

Investigation of Fiber-Inhibitor Systems

A large number of various known and potential urease inhibitors were screened as candidates for treating cellulosic fibers. They became physically attached to the cellulose by sorption from solutions, most practically from aqueous media and then by drying the fiber-based substrates. Few examples of thus treated materials will be given below to illustrate the concept. Practical validity of our approach was evaluated with the aid of the ammonia emission test described above. Some experiments involved the use of substances which serve mainly in medical, veterinary and agricultural applications. One of them was for instance acetohydroxamic acid (AHA), a urea analogue, which is used clinically to dissolve struvite stones [15]. The graphs in Fig. 5 show the effect of cellulose fiber-based nonwoven material containing AHA at two different levels, *i.e.* 100 ppm and 200 ppm by weight of the fabric. This inhibitor can quickly form a stable complex with the active site of urease making it inaccessible to urea. As expected, the nonwoven with acetohydroxamic acid proved itself quite a powerful ammonia-control system. It was not clear though why the fabric containing more AHA was slightly less effective than the one with only 100 ppm AHA. Finally, a comment should be made about acetohydroxamic acid regarding its suspected teratogenic properties [16]. Unfortunately, this information may disqualify the inhibitor for use in certain personal hygiene articles.

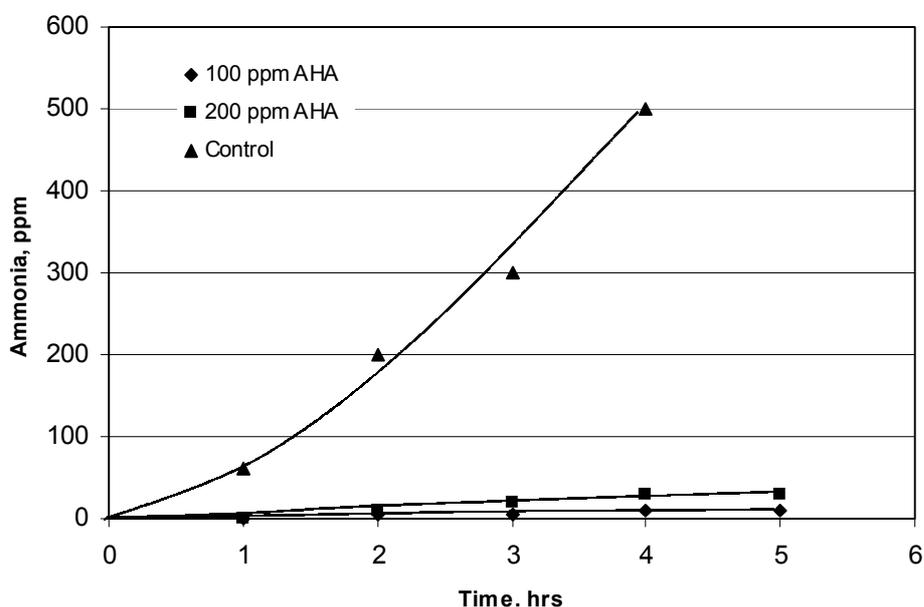


Figure 5. Ammonia emission from Vizorb® 3905 impregnated with acetohydroxamic acid and from untreated control Vizorb® 3905 nonwoven. Each sample (10 g) was insulted with 80 mL urine/urease mixture

Another substance believed to have inhibitory effect on urease is natural saponin extracted from *Yucca schidigera* and used for instance as a supplement to domestic animal diet [17]. *Yucca schidigera* is a plant belonging to the *Agavaceae* family and is native to the deserts of the southwestern U.S. and Baja, Mexico. The saponin extract is available as water-soluble powder. In our experiments its aqueous solution served for treatment of the Vizorb® 3905 nonwoven which was then dried and tested for ammonia control effect. The results shown in Fig. 6 imply that the cellulose-based material containing a small amount of *Yucca schidigera* extract had similar activity as the material modified with acetohydroxamic acid. What remains to be explained again in this study (as in the previous example) is why the effectiveness of the fiber composite with *Yucca* saponin did not correlate with the amount of the applied inhibitor. The *Yucca* extract has been tried as an additive to absorbent articles by other researchers as well [18]. However, the downside of the natural saponin is its rather dark color which may not be desirable in medical and hygiene applications.

There are some reports in the literature suggesting that certain metals such as copper, cadmium, zinc and cobalt [19-21] have an inhibitory effect on urease. The purpose of that research was mainly to find new enzyme inhibitors for biological and medical uses. The authors speculate that these metals can replace nickel atoms in the urease enzyme and thus render it inactive. Aqueous

solutions of various metal salts were used in our work for treating cellulose fiber nonwoven fabric and the obtained samples underwent ammonia emission examination.

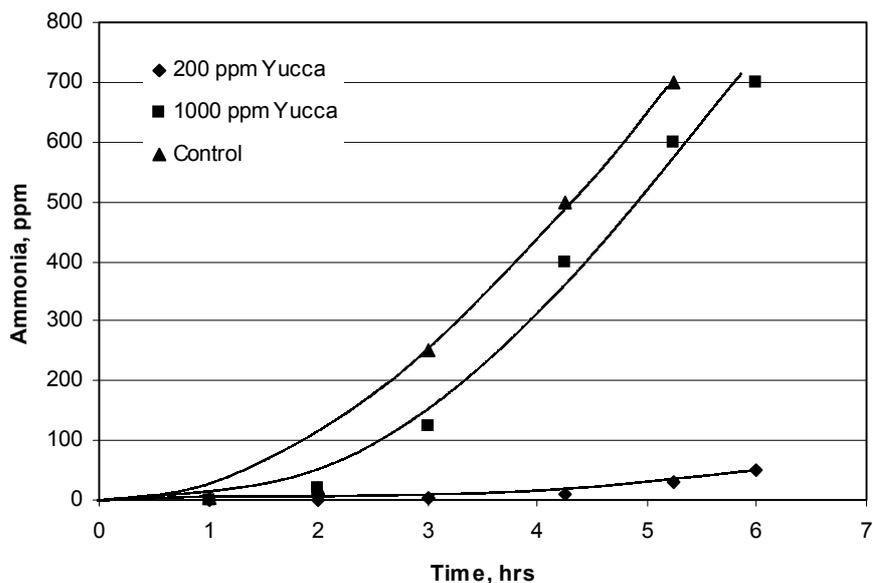


Figure 6. Ammonia emission from Vizorb® 3905 impregnated with *Yucca* extract and from untreated control. Liquid insult volume: 80 mL urine/urease

Some results are illustrated by the graphs in Fig. 7. They compare the inhibitory potency of the tested fibrous materials containing certain metal cations on ammonia emission. Only those polyvalent metals were selected which could be considered generally safe for medical and personal care applications. The data in Fig. 7 suggest that all the experimental materials in this series were able to slow down the generation of NH_2CONH_2 over the time of the experiment. It is interesting to note that the positive results included aluminum and ferric sulfates and these compounds have not been found in the literature search as potent urease inhibitors.

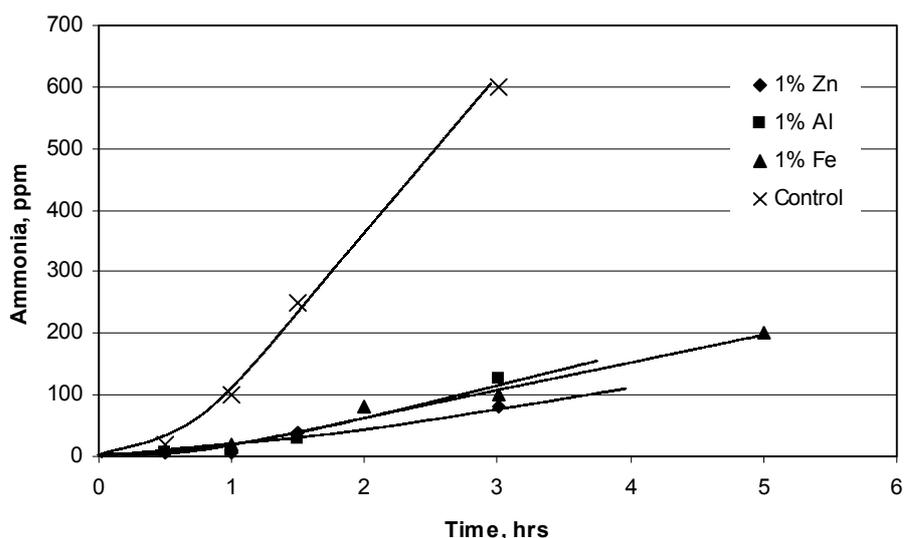


Figure 7. Ammonia generation in Vizorb® 3905 impregnated with a) ZnCl_2 , b) $\text{Al}_2(\text{SO}_4)_3$ and c) $\text{Fe}_2(\text{SO}_4)_3$. Liquid insult volume: 80 mL urine/urease

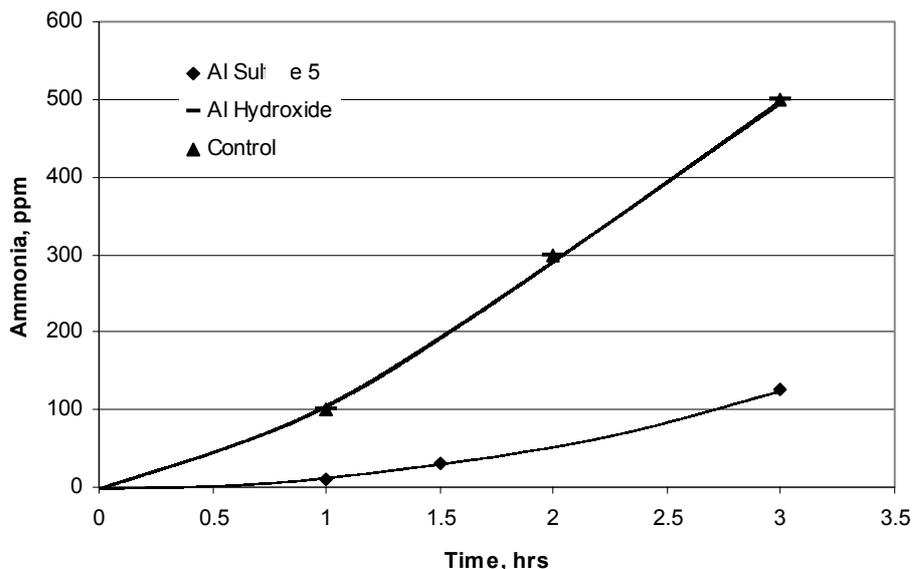


Figure 8. Effect of Vizorb® 3905 impregnated with $\text{Al}_2(\text{SO}_4)_3$ and with $\text{Al}(\text{OH})_3$ on the emission of ammonia after insulating the samples with 100 mL urine/urease

It seems important that a given metal salt have sufficient solubility in order to diffuse to a urease macromolecule and diminish its lytic activity. This can be illustrated in Fig. 8 by comparing the inhibitory activity of soluble aluminum sulfate versus the ineffectiveness of the respective hydroxide deposited on the fibers in the form of insoluble solid. Polyvalent metals are known to have chelating capability which may potentially cause some deformation of the urease macromolecule. As a result, one can speculate that this may be another factor leading to a decrease of the lytic activity of the resultant denatured enzyme.

Some electrolytes may diminish swelling capacity of polyacrylate superabsorbent polymer producing so-called “poison” effect. Therefore, selection of effective metal salts should be done with caution. On the other hand, it is noteworthy that certain polyvalent metal cations may have a beneficial effect on the absorbency characteristics of composites containing superabsorbent particles [22].

Synergy between Fiber with Urease Inhibitor and SAP

As shown earlier in this paper, partially neutralized sodium polyacrylate superabsorbent can bind some amount of ammonia formed as a result of the fermentation of urea. Absorbent cores in disposable hygiene products comprise commonly cellulose fluff blended with SAP powder. The following study illustrates the synergy between the fiber containing urease inhibitor and the superabsorbent polymer. In this case cellulose fluff (Foley Fluffs®, Buckeye) was treated with *N*-(*n*-butyl)thiophosphoric triamide (NBPT, Agrotain), a chemical developed for agricultural application to extend the life of nitrogen contained in urea-based fertilizers. There are numerous compounds designed for this purpose and disclosed in patent literature [13,23]. By and large, some of them are potent urease inhibitors although may have unpleasant smell, are rather sensitive to hydrolysis, oxidation and/or microbial degradation. This instability is a subject of intensive research aimed at better understanding the decomposition mechanism of these compounds and at finding ways of extending their longevity [24].

The results of our experiments illustrated in Fig. 9 suggest that a small addition of sodium polyacrylate superabsorbent to the fiber modified with the inhibitor has beneficial effect on the efficiency of the composite system. However, as the amount of SAP (in this study we always used Favor® 1180 from Degussa) exceeds an optimum, which in this case is about 15%, the ammonia binding capacity of polyacrylate can no longer compensate for a decrease in the urease inhibition activity of the system due to lower content of the modified fiber. For comparison, when the composite containing 50% SAP and 50% fiber without any inhibitor was used in the same series of tests, the concentration of ammonia exceeded 700 ppm (maximum range of the measuring device).

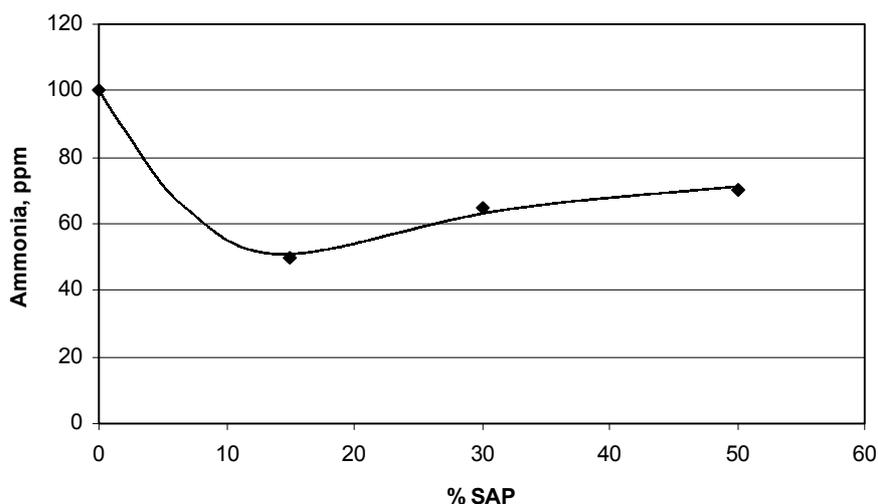


Figure 9. Effect of SAP content in fiber/SAP composite on the amount of ammonia emitted during 20 hours. Cellulosic fiber with 200 ppm NBPT; 10-gram composite samples insulated with 50 mL urine/urease liquid

The New Odor Control Fiber (OCF)

The results of the studies described above became a basis for developing commercial cellulose fiber for controlling the odor of ammonia and other malodors generated from human body fluids. This new, patented product, whose name is Caressa OCF™, is a proprietary technology of Buckeye Technologies Inc. The following main criteria were applied to the evaluation of the OCF product:

- safety, skin wellness,
- odor-control feature,
- absorbency and
- processability.

Caressa OCF™ was subjected to a standard safety study [25], which was carried out by Clinical Research Laboratories Inc. The purpose of this test was to determine any dermal irritation and sensitization potential. In brief, airlaid pads composed of Caressa OCF™ were cut to small squares (1.25 cm by 1.25 cm) and applied to the patch so that the fiber contacted the skin. Prior to the application, the test area of the skin was wiped with alcohol and allowed to dry. Then the patch with Caressa OCF™ remained in direct skin contact for 24 hours. Patches were applied to the same site every second day for a total of 9 applications. The sites were then graded for dermal irritation and sensitization 24 and 48 hours after removal of the samples. The following scoring system served for evaluating observed effects:

- | | |
|----------------------------------------|----|
| - no visible skin reaction | 0 |
| - mild erythema | 1+ |
| - well defined erythema | 2+ |
| - erythema and edema | 3+ |
| - erythema and edema with vesiculation | 4+ |

According to the report [25], based on the test population of 53 subjects, Caressa OCF™ did not demonstrate any potential for eliciting dermal irritation or sensitization.

Caressa OCF™ was tested in the finished adult incontinence products to assess its odor-control performance. The pads were lab-tested using the test described earlier in this paper. Appropriate rectangular pads were cut out of the absorbent core and subjected to the ammonia emission study. The results illustrated in Fig. 10 show significant reduction in the amount of ammonia generated from the experimental pad containing the OCF-based material.

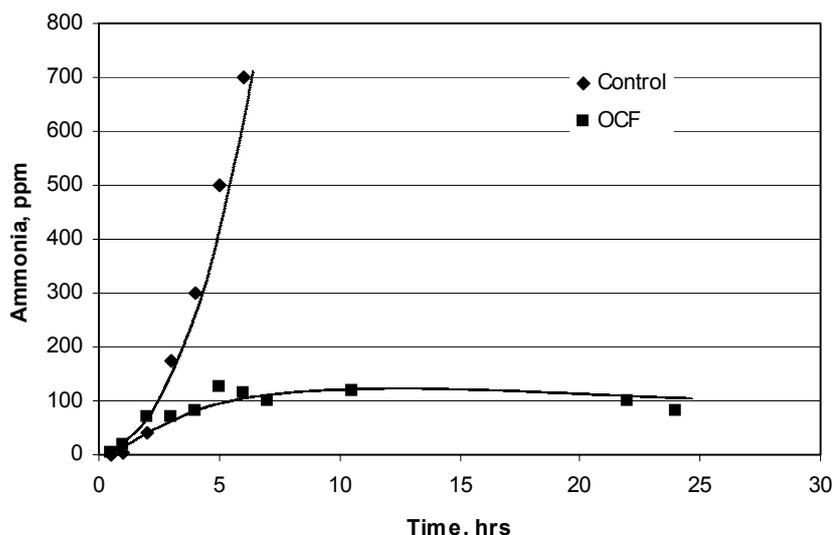


Figure 10. Ammonia control of an experimental AI product versus the effect of a similar product containing regular, untreated fiber

Absorbency capacity is a key property of fiber/SAP composites used in disposable absorbent articles. Therefore, it was important to ensure that the treatment applied to Caressa OCF™ would not have any adverse effect on the retention of liquid by the blend of cellulose fiber and SAP particles. A standard test used in our lab was carried out in the following way: a pre-weighed experimental pad was immersed in saline (aqueous solution of sodium chloride at 0.9% concentration) for 30 minutes under load of 2.8 kPa, then taken out of the liquid, drained for 10 minutes under the same pressure and weighed again. Table 1 shows the results of such experiments carried out with an OCF/SAP composite and with a Foley Fluffs®/SAP control. The data (absorbency capacity) are expressed in grams of liquid retained by one gram sample. As seen from the obtained results the OCF had no adverse effect on the retention of fluid by a blend of cellulose fluff and superabsorbent polymer particles.

Table 1. Absorbency capacity of fiber/SAP pads (basis weight 700 g/m², density 0.17 g/cm³, 70% fiber, 30% SAP)

Fiber grade in fiber/SAP composite	Absorbency capacity, g/g
Foley Fluffs®	9.6
Caressa OCF™	9.6

From the processability standpoint, it is noteworthy that the OCF cellulose pulp sheet can be defibrated with the same disintegration effect and energy consumption as the regular fiber sheet (Foley Fluffs®). This observation is based on our lab experiments and airlaid nonwoven pilot plant trials.

Control of pH and skin wellness

The results of our experiments with composites containing the OCF suggest that the fiber treated with inhibitor should be able to control the pH of skin environment and prevent it from becoming alkaline over time. Maintaining the pH in slightly acid region is beneficial to skin and decreases possibility of irritations and infections. A blend of Caressa OCF™ and SAP (Favor® 1180) was converted into an airlaid pad and insulated with urease-enriched urine in the experimental

equipment shown in Fig. 1. A similar test was conducted for a pad comprising untreated cellulose fiber in lieu of the OCF. In these experiments the pH of urine was measured before insulating the absorbent composite samples and then, after 9 hrs the liquid was extracted from the test specimens and the pH measured again. The results are shown in Table 2.

Table 2. Changes in the pH of urine (80 mL, temp. 37°C) in 10-gram fiber/SAP pads (basis weight 500 g/m², density 0.12 g/cm³, 80% fiber, 20% SAP)

Time, hrs	Caressa OCF™ / SAP composite	Foley Fluffs® / SAP composite
0	5.2	5.2
9	6.1	8.6

In another series of tests the pH readings were taken directly on the surface of the wetted samples. The graphs on Fig. 11 and 12 compare the changes of the surface pH in the samples containing the inhibitor-treated cellulose versus the control samples with regular cellulosic fibers. In these experiments various amounts of fiber/SAP ratios were used in the test samples. Consequently, due to varied capacities of the 10-gram samples different amounts of liquid with urease were used for insults, depending on the content of SAP, i.e. 40 mL for 0% SAP, 48 mL for 10% SAP, 60 mL for 20% SAP and 64 mL for 30% SAP.

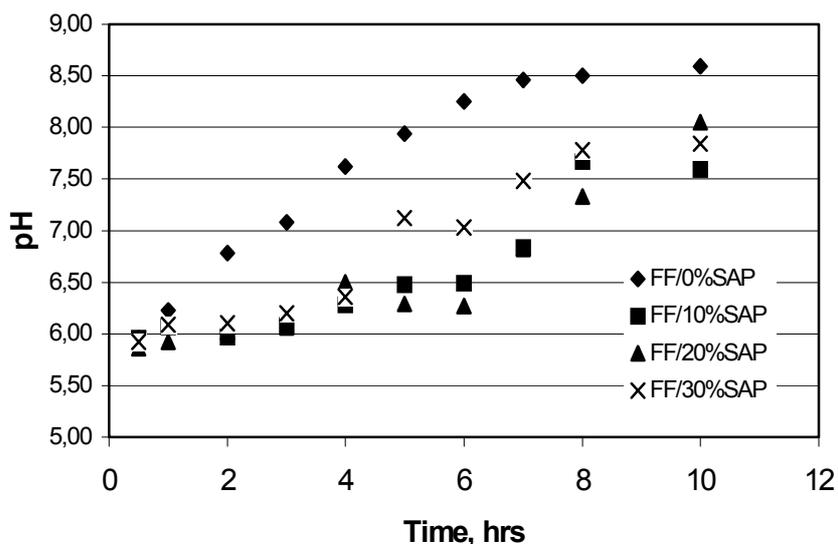


Figure 11. Changes of the sample surface pH in absorbent pads composed of regular cellulose, Foley Fluffs® (FF), and SAP (basis weight 250 g/m², density 0.12 g/cm³)

By and large, the data exhibited in Table 1 as well as in Fig. 11 and 12 clearly show the potential benefit of the modified cellulose when applied to hygienic and medical devices for use in contact with human skin.

As expected, SAP particles in the composites containing regular cellulose fluff were able to slow down to some extent the increase of the pH over time due to the presence of free carboxyl groups in the crosslinked polyacrylic acid salt. Small part of the total amount of alkaline ammonia generated during the experiment could be thus neutralized. However, eventually the pH on the surface of the samples rose above the neutral point. On the contrary, in the series of tests with composites containing the Caressa OCF™ the pH on the surface of the pads never exceeded 6.5.

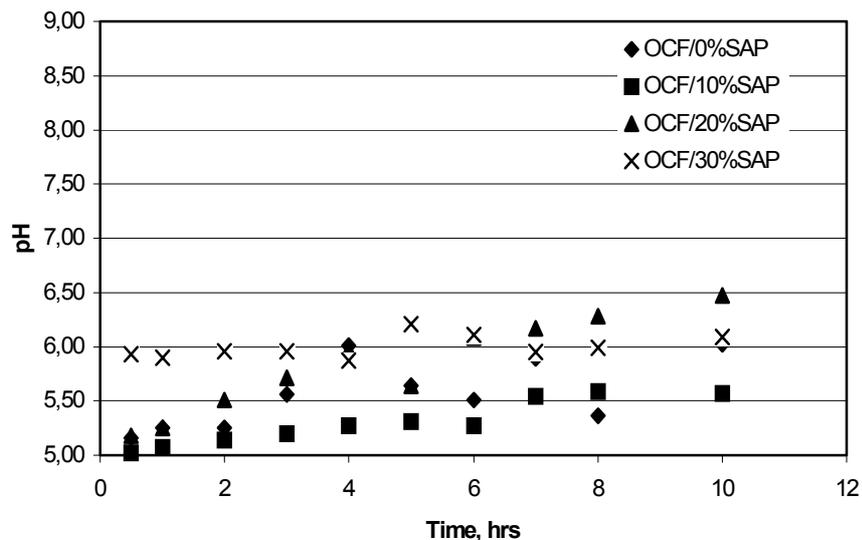


Figure 12. Changes of the sample surface pH in absorbent pads composed of Caressa OCF™ and SAP (basis weight 250 g/m², density 0.12 g/cm³)

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