Enzymatic Debriding Agents: An Evaluation of the Medical Literature

Robert G. Smith, DPM, MSc, RPh, CPed

Although debridement is an essential part of wound care, information to guide evidence-based decisions is limited in the literature. Assuming studies to ascertain the effectiveness of pharmaceutically based enzymatic debridement products are more prolific than studies using nonpharmaceutical debridement options, a literature review was conducted to provide an evidence base to justify current wound care practice. Information on collagenase- and papain-urea-based products was reviewed with emphasis on their functional components, mechanisms of action, and patient considerations. The Medline Database, Cochrane Database of Systematic Reviews, InfoPOEMs, Institute for Clinical Systems Improvement, National Guideline Clearinghouse, and Primary Care Clinical Practice Guidelines were searched for documents published between 1967 and 2007 using the following terms: enzymatic debridement, collagenase, papain-urea, papain-urea chlorophyllin copper complex, wounds, and diabetic foot wounds. Sixteen of 44 relevant citations obtained fit the established criteria for readability, accuracy, reliability and validity of information. Four of the 16 studies included a control treatment, the external validity of 13 studies was limited due to small sample size, and only four studies reported a statistically significant difference in treatment outcome. Predicted bias and publication bias were common. Of the studies detailed herein, three qualified as A level, 13 qualified as B level, and none were considered to provide C level evidence. Although clinicians can glean practical information from the homogenized findings regarding patient demographics, wound type, and therapeutic goals, future studies designed to meet the criteria of level A evidence are needed to provide evidence for the use of enzymatic debridement agents.

KEYWORDS: enzymatic, debridement, collagenase, papain-urea, wound

Dr. Smith is a podiatrist and pharmacist, Shoe String Podiatry, Ormond Beach, Fla. Please address correspondence to: Robert G. Smith, DPM, MSc, RPh, CPed, Shoe String Podiatry, 723 Lucerne Circle, Ormond Beach, FL 32174; email: Robert.Smith@FHMD.org.
the following: clinician experience and skill, cost of the interventional method, the presence of exudate or infection, tissue selectivity, and a patient’s tolerance to a particular intervention. In addition, the most appropriate method can be selected following critical appraisal of the evidence in the literature.

The conventional and customary practice of wound debridement as described in the literature is based on tradition and anecdotal experience, often referred to as best practice approach. This concept is supported by Ryan et al’s observation that evidence-based wound care is still in the developmental stages and the information available has been left to interpretation. From this observation, it can be inferred that the selection of a particular debridement method is generally based on interpretive evaluation rather than evidence-based medicine. Further, Bradley et al assert that compelling clinical evidence comparing one method of debridement with another is scant. Thus, clinical evidence-based information specific to debridement methods is not easy to find. In addition, Beitz has identified the ominous problems related to conducting randomized controlled trials comparing debridement methods — ie, investigation of this nature may create substantial ethical dilemmas for researchers.

The purpose of this literature review is to evaluate the validity and usefulness of enzymatic debridement and provide an evidence base to justify current practice. Enzymatic debridement was chosen not because this method is more effective or preferred compared to other options but because it was presumed that these pharmaceutical products should be associated with a myriad of readily available data from the literature. To serve as a foundation, enzyme products commercially available in the US are reviewed emphasizing functional components, mechanisms of action, and patient considerations. Relevant literature is defined with regards to search determinants and clinimetric definitions, and results are integrated for assessment.

Enzymatic Debridement Agents

Enzymatic debridement is a highly selective method of wound debridement that uses naturally occurring proteolytic enzymes manufactured by the pharmaceutical and healthcare industry specifically for eliminating devitalized tissue. Topical application of exogenous enzymes to the wound surface breaks down necrotic tissue. To allow maximum enzymatic function, a good delivery system, a prolonged period of enzyme activity, and the correct wound environment are required. A review of the literature by Bolton and Fattu found that enzymatic debriding agents are typically used in conjunction with moist wound healing and serve as adjuncts to the autolytic debridement process. Falanga’s clinical practice review underscores a paradigm shift — ie, that endpoints for determining success with enzymatic products include debridement as well as wound closure. This new attitude may have been the motivation for the Centers for Medicare and Medicaid Service (CMS) announcement that as of January 1, 2008, all enzymatic debridement products without an approved Food and Drug Administration application will be removed from the 2008 Formulary Reference File. This action by CMS reduced therapeutic options in the Dermatological Wound Care Agent Class and had an impact on enzymatic debridement options. Now reimbursement for commercially available products is limited to collagenase, papain/urea, and papain/urea/chlorophyllin complex.

Collagenase-based products. Collagenase is a water-soluble proteinase indicated for the enzymatic debridement of necrotic tissue in the treatment of...
severe burns and dermal (including decubitus) ulcers. It is derived from Clostridium histolyticum, a member of the metallopeptidase family.\textsuperscript{16} Collagenase specifically hydrolyzes peptide bonds and digests all triple helical collagen and will not degrade any other proteins lacking the triple helix — a unique feature of bacterial collagenase because no other available protease can digest collagen.\textsuperscript{17} The enzyme has been shown in in vitro and in vivo studies to liquefy necrotic tissue without damaging granulation tissue. Collagenase digests the lower portion of an eschar, working from the bottom up; thus, it appears to work more slowly than other debriders.\textsuperscript{7} In both in vitro and in vivo investigations, collagenase has been shown to be gentle to viable cells and may promote angiogenesis and epithelialization.\textsuperscript{18} It is believed that collagenase-based enzymatic agents may remove substrates necessary for bacterial proliferation or may afford antibodies, leukocytes, and antibiotics better access to the infected area; additional information is available that is beyond the scope of this article’s intent. Clinical advantages include selective removal of dead tissue, painless application, enhanced proliferation and migration of keratinocytes, and minimal blood loss.

Collagenase ointment may be applied directly to the wound or to a sterile gauze pad applied to the wound and properly secured. The substance is stable at physiological pH (6.8 to 7.4) and has been found to have optimal activity at a pH of 6 to 8.7.\textsuperscript{16} The enzyme is inactive below pH 5 and at pH 8.5 or greater but enzymatic activity can be restored when the pH is returned to 7.4.\textsuperscript{16} Furthermore, collagenase can be destroyed by other proteolytic enzymes or by temperatures exceeding 56° C.\textsuperscript{16} Collagenase enzymatic activity is adversely affected by detergents, heavy metal ions (eg, mercury, zinc, or silver), and povidone iodine antiseptic solutions.\textsuperscript{7,16} However, hydrogen peroxide, Dakin’s solution, 0.9% sodium chloride solution, polymyxin B, and bacitracin do not interfere with its activity.\textsuperscript{7,16,18}

**Papain-based products.** Papain is a nonspecific proteolytic enzyme derived from the fruit of the papaya tree (Carica papaya).\textsuperscript{19-21} Papain breaks down fibrinous material in necrotic tissue; it requires the presence of sulphydryl groups (eg, cysteine) found in such tissue to stimulate activity.\textsuperscript{5,11} Papain does not digest collagen.\textsuperscript{21} The addition of urea helps expose the activators of papain in necrotic tissue by altering the three-dimensional structure of proteins and disrupting hydrogen bonding.\textsuperscript{15,17} In human comparative studies,\textsuperscript{7} the combination of papain and urea has been found to be approximately twice as effective at digesting protein compared with papain alone. Ayello and Cuddigan\textsuperscript{1} published practice guidelines reviewing collected research that suggests papain-urea affects the biologic activity of recombinant human platelet-derived growth factor BB. They note that papain breaks down proteins containing cysteine and remind clinicians that growth factors contain cysteine residues. Thus, they dispute manufacturer’s claims that papain-urea is harmless to viable tissue. Although shown to be generally well tolerated and nonirritating according to one practice guideline reviewing the manufacturer literature,\textsuperscript{15,18} papain-urea preparations produce more exudate when digesting eschar, which may irritate the surrounding skin.\textsuperscript{7} These products should be applied daily with a moisture-retentive dressing; this type of dressing with an adhesive border or secondary dressing allows patients to maintain activity without disturbing the wound.\textsuperscript{18} In addition, papain use is known to produce an inflammatory response in vivo and case studies/series have shown that some patients experience considerable pain with its use.\textsuperscript{21,22} Per clinical opinion, the addition of chlorophyllin (a substance that prevents formation of agglutinated erythrocytes) has been found to reduce pain.\textsuperscript{15} Hydrogen peroxide solution, as well as salts of heavy metals such as lead, silver, and mercury may inactivate papain.\textsuperscript{7,18-20}

**Papain-urea-chlorophyllin copper complex.** Chlorophyllin is an antiagglutinin and prevents the formation of agglutinated erythrocytes.\textsuperscript{18} Sodium copper chlorophyllin is an historically established wound healing product.\textsuperscript{20} The inclusion of chlorophyllin copper complex sodium with papain-urea allows a product’s continuous use for as long as necessary to help produce and then maintain a clean wound base, promote healing,\textsuperscript{4} and reduce odor. Papain-urea chlorophyllin copper complex should be applied daily and covered with an appropriate dressing. Brett’s\textsuperscript{20} review of clinical study results suggests that papain-urea chlorophyllin copper complex’s
proteolytic action thoroughly cleanses lesions of all necrotic tissue debris and then maintains optimal circulation; affected tissue benefits from both hematological and nutritive elements.

**Method**

**Literature search strategy.** To ensure the information found in the enzymatic debridement literature would be representative of clinical practice, Medline Database, Cochrane Database of Systematic Reviews, InfoPOEMs, Institute for Clinical Systems Improvement, National Guideline Clearinghouse, and Primary Care Clinical Practice Guidelines were searched for primary sources. A search profile was compiled using the following subject terms: enzymatic debridement, collagenase, papain-urea, papain-urea chlorophyllin copper complex, wounds, and diabetic foot wounds. These key words and phrases were used to perform Boolean logic electronic searches to identify relative literature citations. The search covered primary citations published between 1967 and 2007 (spanning 40 years). A manual review of the citations’ references lists and bibliographies was performed to extract additional information that might lead to further material for this review. Conference or proceeding presentations were excluded from this review.

**Clinimetric property terms.** Clinimetric properties used to examine and evaluate the evidence include readability, accuracy, reliability, and validity. Readability refers to the participant’s understanding of the text. Accuracy is defined as careful, exact, and free from errors; therefore, accuracy is the state of being accurate or having precision. Reliability is the degree to which a measure can be reproduced. Reliability is a necessary condition for validity but is not a sufficient condition for validity. Inter-rater reliability is an evaluation of the reproducibility of a measure between observers.

Validity is the extent to which an instrument measures what it is intended to measure. Validity may be classified in six ways: face validity, content validity, criterion-related validity, concurrent validity, predictive validity, and construct validity. Face validity is validity taken at face value. Face validity is the weakest form of measurement. Content validity draws an inference from test scores to a large domain and relates to sample-population representativeness. Content validity often applies to questionnaires and inventories that comprise an instrument and how well, when considered together, they address the issues.

Content validity provides evidence when content experts are in agreement. Criterion-related validity is determined by benchmarking the new measure against a gold standard already in existence. Riegelman and Hirsh describe a gold standard as the criterion used to unequivocally define the presence of a condition or disease under study. Predictive validity is used to predict a future criterion score by establishing the outcome of the target. Concurrent validity is established when two measurements are taken at relatively the same time. This type of validity is used to determine whether a new target test is more efficient than the existing “gold standard.” Construct validity examines the agreement between the measure and the theory of what the tool should be measuring in the instructions or items included. Construct validity is also known as theoretical construct because it draws an inference from test scores. Test bias is a major threat to construct validity.

**Defining levels of evidence.** The ABC rating scale for evidence-based clinical reviews recommended and defined by Siwek et al. was used to describe the levels of evidence for each publication obtained in the literature search. Level A applies to randomized controlled trials/meta-analyses found when high quality randomized controlled trials consider all important patient outcomes. These trials employ quantitative systematic review using comprehensive search strategies and are considered strong evidence. Level B designates a well designed, nonrandomized clinical trial and reflects search strategies involving nonquantitative systematic review with well-substantiated conclusions. Reviews comprising B-rated evidence include lower quality randomized controlled trials, clinical cohort studies, and case-controlled studies with nonbiased selection of study participants and consistent findings. Other evidence (historical, uncontrolled studies and well designed epidemiologic studies with compelling findings) may qualify for a B rating as long as it is of high quality.

A level C or suggestive evidence rating is assigned to publications presenting consensus viewpoints,
expert opinions, or panel interpretation of uncontrolled or observational studies on a specific subject. Expert opinion must not be confused with personal experience that also may be referred to as *eminence-based medicine*. Siwek et al. emphasize that each rating is applied to a single article, not to the entire body of evidence that exists on a topic.

### Results

Of the 44 citations found in the National Library of Medicine and the National Institutes of Health PubMed-Medline databases, 28 were review articles. The Cochrane Database and the National Guideline Clearinghouse include either a systematic review with explicit criteria or evidence-based guidelines. Searching the Cochrane Database using the phrase *enzymatic debridement* yielded 13 documents. The National Guideline Clearinghouse was searched using the stipulated criteria and the phrase *enzymatic ulcer debridement*, yielding six summaries—three do not specifically provide supporting evidence, one is denoted as level B evidence, and two are classified as level C evidence. The same search phrase used for InfoPOEMs, the Institute for Clinical Systems Improvement, and the Primary Care Clinical Practice Guidelines databases yielded no results.

### Enzymatic Debridement: Study Summaries (see Table 1)

**Lee and Ambrus.** Lee and Ambrus' double-blinded randomized comparison trial of collagenase

<table>
<thead>
<tr>
<th>Authors (year published)</th>
<th>Treatment</th>
<th>Study Design</th>
<th>Reported Outcomes/Results</th>
<th>Level of Evidence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lee and Ambrus (1975)</td>
<td>Collagenase</td>
<td>Double-blind randomized</td>
<td>14/17 ulcers improved No statistical difference</td>
<td>B</td>
</tr>
<tr>
<td>Parish and Collins (1979)</td>
<td>Collagenase</td>
<td>Observational case series</td>
<td>13/21 ulcers improved No statistical difference</td>
<td>B</td>
</tr>
<tr>
<td>Rao et al (1975)</td>
<td>Collagenase</td>
<td>Randomized observational case series</td>
<td>80% wound closure Fewer days to treat</td>
<td>B</td>
</tr>
<tr>
<td>Vetra and Whittaker (1975)</td>
<td>Collagenase</td>
<td>Randomized matched paired</td>
<td>Efficacy/economic analysis: complete wound healing</td>
<td>A</td>
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<tr>
<td>Hansbrough et al (1995)</td>
<td>Collagenase</td>
<td>Prospective randomized blinded</td>
<td>No statistical difference No efficacy difference</td>
<td>B</td>
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<tr>
<td>Muller et al (2001)</td>
<td>Collagenase</td>
<td>Prospective randomized blinded</td>
<td>No statistical difference</td>
<td>B</td>
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<tr>
<td>Pullen et al (2002)</td>
<td>Collagenase</td>
<td>Prospective randomized blinded</td>
<td>No efficacy difference</td>
<td>B</td>
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<tr>
<td>Konig (2005)</td>
<td>Collagenase</td>
<td>Prospective randomized blinded</td>
<td>No statistical difference</td>
<td>B</td>
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<tr>
<td>Gasser (2006)</td>
<td>Papain-urea-chlorophyllin</td>
<td>Observational case series</td>
<td>27/30 good results</td>
<td>B</td>
</tr>
<tr>
<td>Morrison and Casall (1957)</td>
<td>Papain-urea-chlorophyllin</td>
<td>Observational case series</td>
<td>Results inconclusive</td>
<td>B</td>
</tr>
<tr>
<td>Katz et al (1956)</td>
<td>Papain-urea-chlorophyllin</td>
<td>Observational case series</td>
<td>Effective debrider No significant in wound area between agents</td>
<td>A</td>
</tr>
<tr>
<td>Burke and Golden (1958)</td>
<td>Papain-urea-chlorophyllin</td>
<td>Observational case series</td>
<td>No statistical difference</td>
<td>B</td>
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<tr>
<td>Alvarez et al (2002)</td>
<td>Papain-urea-chlorophyllin</td>
<td>Observational case series</td>
<td>No significant in wound area between agents</td>
<td>A</td>
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</table>
against a placebo (heat-inactivated enzyme) to determine the effectiveness in pressure ulcers is one of the first reports to show a statistically significant difference between collagenase and placebo treatment. Study participants included three men and eight women with advanced dermal wounds including lower leg stasis ulcers and decubitus ulcers involving the sacrum, coccyx, trochanter, heel, elbow, ankle, knee, and foot (the authors presented no inclusion or exclusion criteria). Each patient's ulcer was treated daily with either collagenase or a placebo. Of the 28 ulcers studied, 17 were treated with collagenase and 11 with heat-inactivated collagenase placebo. The outcome goal for this investigation was "progress to healing" as weekly wound volume measurement up to 30 days. Two diameters of the ulcer were measured and color photographs of the lesion were taken. A volume mold was created using the same material used to prepare impressions of oral lesions; casts of this material dehydrate and fall apart within 4 days. Wound volume was determined by water displacement.

Progress — defined by the authors using wound measurement and the presence/absence of odor, pus, inflammation, and necrosis — was considered good in 14 and poor in three of the collagenase-treated ulcers. The pressure ulcers in both groups increased in size with a predicted bias toward enzymatic treatment — if the increase in volume was associated with the appearance of healthy granulation tissue, the overall evaluation was rated "good."

Despite the authors' claim that overall results were based on objective and subjective criteria, only the terms "good" and "poor" were used to express overall evaluation, limiting the ability to delineate and apply the findings with confidence. Also, this study makes no mention of the statistical tools or analysis used to support the authors' statement that baseline wound volume was significantly different between groups. Randomization and blinding methods are not described. Patient withdrawals are mentioned without any explanation. On the other hand, this is the first study to describe the use of a control group on this subject. Although elements of construct validity appear within this investigation, possible test bias on the part of these authors compromises the validity of the findings.

Parish and Collins. Using an observational case series, Parish and Collins compared use of a collagenase enzyme preparation against sugar and egg whites as a placebo/control applied after saline wash on 34 pressure ulcers in 17 patients treated in a community nursing home. Eleven ulcers in five patients were treated with collagenase and nine ulcers in five patients were treated with the sugar-and-egg-white control. The outcome goal of this study was healing as measured by mean wound size surface area at the end of 4 weeks. Elements of criterion validity were observed with this investigation because collagenase results were compared to a "gold standard" of that time. Two of the five patients in the collagenase group improved at the end of the study and none of the five patients in the sugar-and-egg-white treatment arm improved, leading authors to conclude that collagenase can be helpful in the treatment of decubitus ulcers. However, no statistical difference between the groups for ulcer size was detected. The male-to-female ratio of the study population is not presented, nor do the authors describe the randomization procedure or explain patient exclusion criteria although they state a control method was employed. No side effects or withdrawals were reported for patients in either group.

Rao et al. Rao et al authored a descriptive, randomized study of the therapeutic benefits of collagenase ointment in 24 patients (16 women, eight men) with pressure ulcers in need of debridement. All ulcers underwent standardized saline wound cleaning and collagenase ointment was applied to the lesions daily for an average of 3 weeks until endpoint, defined as sufficient debridement to achieve abundant granulation tissue. Degrees of odor, pus, necrosis, inflammation, granulation tissue, and subjective pain were rated using nonparametric scales (absent = 0, mild = 1, moderate = 2, and severe = 3). Therapeutic responses were rated using the descriptive terms excellent, good, fair, or poor. Little or no change in severity of inflammation occurred until the end of the third week of therapy; improvement was statistically significant at the 1% level ($P < 0.001$). Also, the rate of increase in granulation over the 3-week period was found to be statistically significant ($P < 0.005$). Wound size
decreased in 13 cases and remained unchanged and increased in size in four cases, respectively, suggesting that collagenase is an excellent addition to total therapeutic and nursing management of decubitus ulcers.

However, although a statistical analysis section was provided, the authors did not define the statistical test used to support their findings. Also, these authors detail difficulties encountered when conducting controlled studies of therapeutic agents for decubitus ulcers that may have compromised the results — ie, variability in wound demographics and challenges in measuring slow-healing wounds, estimating fluctuations in the underlying comorbidities, and addressing patient nutritional status.

Vetra and Whittaker. Vetra and Whittaker’s observational case series investigated the effectiveness combining hydrotherapy and collagenase in 140 wounds of varying etiology (arteriosclerosis, burns, decubitus, diabetic, gangrene, perforating, postsurgical, pressure, traumatic, and vascular). The objective was time to total healing (per wound measurement). Within 37 days, 80% of the wounds completely closed. The authors concluded that the combined use of hydrotherapy and collagenase improves debridement and the subsequent closure of ulcerative lesions.

The authors acknowledged they were more concerned with the time required for total healing and the final clinical response than with the early stages of granulation and initial epithelialization. In the discussion of this investigation, no mention is made of randomization, control group, or evaluation against accepted standard of care therapy for the time.

Hansbrough et al. Hansbrough et al conducted a randomized, multicenter (eight) comparison of the efficacy of collagenase applied with polymyxin B sulfate/bacitracin power versus a silver sulfadiazine cream control. Participants included 79 patients (64 men, 15 women) with partial-thickness burns (flame, flash, scald, or other) wounds randomly assigned to either the treatment or control group. Therapeutic endpoints were defined as time to clean wound bed or time to heal by mean wound size as measured by a designated observer. These authors found when paired treatment sites were compared within each patient, proportionally more of the sites treated with collagenase “cleaned” and healed faster than similar wounds treated with silver sulfadiazine. As a secondary finding, there was no evidence of increased pain associated with the use of collagenase treatment assessed in a subset of 19 patients.

These authors applied rigorous statistical analysis to their data and incorporated previous published data as a pilot foundation for their investigation. Also, they compared collagenase to the "gold standard" (ie, silver sulfadiazine). Finally, although the patient population was not highly randomized, two anatomically similar partial-thickness wounds provided the basis for comparison. These authors utilized concurrent validity to determine if collagenase treatment is more efficacious than sulfadiazine for thermal injuries for two therapeutic endpoints.

Muller et al. Muller et al’s prospective, randomized trial of 24 female hospitalized patients with grade IV pressure ulcers on their heels compared collagenase (n = 12) and hydrocolloid (n = 12) products. The therapeutic outcome goals for this investigation were total epithelialization and cost of therapy. Treatment efficacy was assessed by size and depth of the ulcer, signs of inflammation, formation of new necrotic tissue, presence of granulation tissue, and epithelialization. The robustness of the results was tested with several sensitivity analyses, which revealed that the collagenase was more cost effective than the hydrocolloid, primarily due to a shorter duration of treatment that resulted in reduced personnel costs.

The authors applied rigorous statistical analysis to their findings on effectiveness of product and cost. The study involved comparison to the gold standard (hydrocolloid) for pressure ulcers and randomization and two therapeutic endpoints provided elements of concurrent validity.

Pullen et al. Pullen et al’s randomized, prospective, double-blind trial involved 135 elderly patients with pressure ulcers. Of the 78 patients for whom data were available, 44 were treated with collagenase and 34 with fibrinolysin/DNASE; wound debridement or observed changes in wound environment served as the study endpoint. The statistical evaluation of the change of necrotic wound area showed slightly better (but not statistically significant) results for the collagenase group compared to the fibrinolysin/DNASE group.
A detailed description of randomization is provided and rigorous statistical analysis applied. The use of rigorous statistical analysis facilitates conclusions, ensuring minimal test bias and validating the investigation’s theoretical construct.

Konig et al. Konig et al. conducted a 21-day randomized clinical trial comparing the effectiveness of enzymatic debridement to natural autolytic debridement in chronic leg ulcers. Of the 42 patients enrolled in this investigation, 15 received a moisture-activated polyacrylate dressing (TenderWet 24, Medline Industries, Inc., Mundelein, Ill), and 27 patients received collagenase. Therapeutic outcome goals were reduction of eschar or development of granulation tissue and epithelialization as measured by digital images. Although the polyacrylate dressing appeared to be more efficient in a few cases, the general efficacy of the two treatment approaches appeared to be almost the same (no statistically significant difference) using Wilcoxon matched-pairs test; sectional comparisons between groups using independent pairs U-test failed to show superiority of either product. The authors concluded that enzymatic debridement was not superior to moist debridement in their setting.

The authors acknowledge they did not employ blocked randomization, resulting in unequal numbers in the therapeutic groups. However, these authors employed a gold standard (autolytic debridement); thus, this investigation demonstrates criterion-related validity.

Marazzi et al. Using a large patient database to conduct a retrospective analysis, Marazzi et al investigated the effect of enzymatic debridement with collagenase on both acute and chronic hard-to-heal wounds. Over a 4-year period, the records of 979 patients (adults and children) with hard-to-heal wounds who were treated with collagenase-based ointments were compiled and analyzed. Inferential comparisons, nonparametric tests, and a linear regression model were used to correlate healing times for chronic ulcers with the wound area at the start of treatment.

These authors did not offer a definition of hard-to-heal wounds and no information concerning randomization or controls is provided. Changes in wound characteristics and healing time are offered as therapeutic outcome goals and measured subjectively.

Grasser. In 1940, Grasser published a preliminary report of 58 patients with sloughing wounds treated with an enzyme mixture consisting of papain, triethanolamine, oleic acid, stearic acid, and mineral oil mixed with an activator consisting of an aqueous solution of 5% triethanolamine. This emulsion was applied to wounds after cleaning with normal saline and covered with cellophane or rubber seals daily or every other day. The therapeutic outcome goal was time to achieve clean granulation tissue. Twenty-nine cases were described as old leg ulcers with slough. The average number of enzyme emulsion applications necessary to remove slough and produce a clean wound was two and the average number of treatment days until granulation tissue was present was 6.15.

Wound measurement evaluation involved subjective observational analysis. This investigation was not randomized, controlled, or blinded. Moreover, the statistics provided were rudimentary. Although findings within this investigation established a foundation to provide data for the successful use of papain treatment, Grasser’s effort in researching enzymatic treatment of sloughing wounds does not meet criteria for construct validity. Despite the author’s observations, predicted and test bias must be considered.

Miller. Miller conducted a case series investigation among 39 patients with a wound resistant to previous therapy, over a bony prominence, or caused by pressure or shear to compare the efficacy and safety of papain-urea-chlorophyllin (n = 24 patients) to papain-urea (n = 15). Wounds were cleaned with normal saline before application of either product and covered by a nonadherent primary dressing either once or twice a day. Therapeutic outcomes and goals for this investigation were either complete debridement or complete healing of the treated wound. Twenty-three patients with pressure ulcers, previously resistant to therapy, who used papain-urea-chlorophyllin copper complex sodium therapy were completely healed within 3 months. Interestingly, the authors describe that good digestive action was observed within 2 or 3 days among all 15 patients receiving papain-urea; the formation of granulation tissue was not observed because all 15 patients in the papain-urea group had to discontinue the study because of local irritation.
Exclusion criteria were not specified in this descriptive case series. Wound measurements were evaluated subjectively. All patients in the papain-urea therapeutic arm experienced local inflammatory reactions within 1 to 3 days and had to stop therapy. The concentration of the papain-urea is not specified, making it difficult to apply this observation to other populations. The study is limited by its small size and case design. Additionally, interpreting the data is difficult without considering the possible presence of bias toward the papain-urea treatment group. Although elements of face, predictive, and criterion-validity are present in this investigation, they are influenced by investigator test bias.

**Morrison and Casali.** Morrison and Casali's case series addressed papain-urea-chlorophyllin ointment applied daily under standard dressings as proteolytic therapy for 20 women and 10 men with decubitus ulcers previously resistant to topical therapy. Inclusion criteria specified wounds over a bony prominence caused by pressure or shear, wounds resistant to previous topical antibiotic therapy, and patients between the ages of 50 and 80 years; exclusion criteria were not specified. Therapeutic outcome was rate of complete healing determined by subjective observations. Of the 30 study participants, 27 were completely healed within 2 to 6 weeks with the papain-urea-chlorophyllin ointment.

It was not possible either to include a parallel control series or to take microscopic sections to study the effect of the drug on capillary and lymphatic circulation. Results were reported as superior and significant compared to any previous therapies but previous therapies were not described. Study limitations include small case design, no provision of baseline characteristics for the case series group, and no assessment of clinical benefit of wound closure after the treatment period. The authors attempted to use construct validity to produce agreement between their outcome measures and the theoretical outcome but the limitations did not allow the theoretical construct to be completely validated.

**Katz et al.** Using a descriptive case series format, Katz et al studied use of papain-urea-chlorophyllin ointment for eight categories of lesions in 24 men and 26 women. The papain-urea-chlorophyllin ointment was applied once or twice a day covered by a sterile dressing depending on necrotic material present. Inclusion criteria were the presence of wounds (carbuncles, decubitus ulcers, diabetic ulcers, varicose ulcers, infected wounds, post-phlebitic ulcers, arteriosclerotic ulcers) and burns. Therapeutic outcome goals were duration of therapy and changes in wound characteristics based on subjective evaluation. Healing was described nonparametrically using good, fair, and poor: good indicated a decrease in purulent material, cleaner granulation tissue, and increased rate of epithelialization; fair was a slight but definite improvement; and poor was defined as no change or actual worsening of the lesion. Among the 50 participants, healing in 37 cases
(74%) was classified as good, six (12%) as fair, and seven (14%) as poor. Of the four patients presenting with diabetic ulcers, two had a poor outcome, one was good, and one was fair. Papain-urea-chlorophyllin ointment was described as a successful chemical debriding agent and had no effect on viable tissue.

Construct validation would have been enhanced had the authors provided wound measurements for increased objectivity. Outcome data and conclusion may have been more convincing if statistics were explained in detail.

**Carter.** Carter’s case series investigated 37 patients with wounds resistant to previous antibacterial therapy who were treated with papain-urea-chlorophyllin ointment. Therapeutic outcome goals included duration of therapy to obtain clean granulation tissue and number of days to achieve complete wound healing. Inclusion criteria were presence of decubitus ulcers (28 patients), leg ulcers (12 patients), and pressure ulcers (two patients); no exclusion criteria were noted. All ulcers had been treated previously with antibiotic or antibacterial ointments (including vitamin ointment and ichthyol for 24 days to 3 years. Wounds were treated with this papain-urea-chlorophyllin ointment under gauze pads changed once (most patients) or twice daily depending on amount of exudate. The author infers from the findings that the time required for complete healing using the papain-urea-chlorophyllin ointment was half of what had come to be expected from traditional antibacterial treatment for similar patients presenting with similar ulcers.

Because no control was in place, results on healing rates lack comparability. The author acknowledges that data are inconclusive and intended only to provide an instructional tool.

**Burke and Golden.** Burke and Golden conducted a prospective, randomized, controlled efficacy trial between collagenase and papain-urea formulations to evaluate and compare the ability of two chemical debridement ointments to remove devitalized tissue in pressure ulcers. After completing either a 1- or 2-week screening period to stabilize their wounds, 28 nursing home patients with pressure ulcers from three centers were randomly assigned by computer to receive either collagenase (n = 12) or papain-urea formulations (n = 14); two patients withdrew due to unrelated issues. Criteria for inclusion were wounds caused by pressure, shear, friction, or excessive moisture in mobility-compromised individuals; full- or partial-thickness wounds; wounds in need of debridement based on the investigator’s opinion; wounds with nonviable tissue attached to the base of wound; and an ankle brachial index >0.75 for foot wounds. Patients were excluded if infection, cellulitis, inadequate nutrition, and uncontrolled diabetes or unstable comorbidities were present. Therapeutic outcome endpoints included reduction of nonviable tissue and degree of granulation tissue as determined by clinical evaluation. The product was applied (approximately 2 mm thick) once daily using a tongue depressor over the entire wound or surface of nonviable tissue. Reapplication (up to two applications per day) was provided only if the dressing came off or became soiled. Data analysis revealed that underlying pathologic condition,” negatively influencing criterion validation of this investigation.

These authors state the study was entirely clinical and objective but only 18 patients are represented in photographs. Also, wound dimensions or statistics are not presented. The duration of initial therapy and treatment therapy are listed but not discussed sufficiently. Detailed patient discontinuation information is not provided. The authors assert that their findings parallel previously published data, justifying widespread use of papain urea chlorophyllin ointment. The mention of the parallel studies and declaration of clinical objectivity is an attempt to validate (criterion-related) the investigation, creating potential bias in favor of the papain-urea-chlorophyllin intervention. The authors claim significant savings that were compatible with the patients’ welfare but present no economic data to substantiate this observation.

**Alvarez et al.** Alvarez et al conducted a prospective, randomized, controlled efficacy trial between collagenase and papain-urea formulations to evaluate and compare the ability of two chemical debridement ointments to remove devitalized tissue in pressure ulcers. After completing either a 1- or 2-week screening period to stabilize their wounds, 28 nursing home patients with pressure ulcers from three centers were randomly assigned by computer to receive either collagenase (n = 12) or papain-urea formulations (n = 14); two patients withdrew due to unrelated issues. Criteria for inclusion were wounds caused by pressure, shear, friction, or excessive moisture in mobility-compromised individuals; full- or partial-thickness wounds; wounds in need of debridement based on the investigator’s opinion; wounds with nonviable tissue attached to the base of wound; and an ankle brachial index >0.75 for foot wounds. Patients were excluded if infection, cellulitis, inadequate nutrition, and uncontrolled diabetes or unstable comorbidities were present. Therapeutic outcome endpoints included reduction of nonviable tissue and degree of granulation tissue as determined by clinical evaluation. The product was applied (approximately 2 mm thick) once daily using a tongue depressor over the entire wound or surface of nonviable tissue. Reapplication (up to two applications per day) was provided only if the dressing came off or became soiled. Data analysis revealed that
papain-urea proved to be more effective for pressure ulcer debridement and appeared to be more effective in promoting granulation tissue than collagenase ($P < 0.0167$). Also, papain-urea significantly reduced the area of necrotic tissue as measured by planimetry techniques at 4 weeks compared to collagenase in pressure ulcers requiring conservative debridement ($P < 0.0167$). No statistically significant difference in the quantity of resident bacteria between treatment regimens was found.

The relatively small patient population prohibited rendering strong scientific conclusions even though the papain-urea debriding ointment exhibited advantages over the collagenase debriding ointment. Several limitations affected outcomes: the study was not blinded to either investigators or patients, complete wound closure or the incidence of complete healing was not reported, and because complete wound closure was not assessed after 4 weeks, the durability of effect as well as a surveillance of adverse effects from either product could not be measured.

Wight and Shi. Wight and Shi offer a historical review of a papain-urea ointment (Accuzyme®, Healthpoint, Ltd, Fort Worth, Tex) that discusses the physical and chemical properties, mode of action for a papain-urea debriding agent, and clinical experience with papain-urea debriding preparations. The review includes information from Carter, Morrison and Casali, and Burke and Golden and refers to the enzymatic debriding intervening agent as “papain-urea debriding preparations” instead of papain-urea-chlorophyllin, which was the product investigated. Wight and Shi attempt to use content validity in their review by illustrating successful outcomes with papain-urea debriding ointment — paraphrasing implies an element of publication bias toward the proprietary product and perhaps a reflection of commercial interest.

Discussion

Clinical evidence search. Many review articles describe the art and science of enzymatic debridement. Only a few citations found within the published medical literature actually evaluate enzymatic debridement using control subjects and even fewer
describe actual randomized controlled trials. This suggests that few clinical studies have been conducted since Bradley et al’s 1999 review was published. Relevant clinical literature evaluating collagenase and papain-urea combinations is limited. Comparing published articles to provide evidence ensures that minimum criteria for both research application and clinical practice are met. Comparisons should articulate the effect of study methods, as well as the therapeutic goal (eg, debridement, healing rate) on results.

Systematic evaluation is essential when approaching clinical questions to ensure they are assessed on an even plane. A wound care specialist can apply the information gleaned from related literature sources to produce specific workable patient solutions. Four categories are common to the research findings and can be used to evaluate the 16 clinical practice articles found in this literature search: patient populations, wound types, wound measurements before and after inventions, and outcome goals.

**Patient population.** Of the 16 studies discussed, 12 provide both gender and age demographics and can facilitate comparison to a practitioner’s particular patient population. The number of subjects is crucial to accurate statistical analysis of the data. Many of the studies published involve fewer than 100 subjects; this is usually inadequate to provide reliable statistics because a rule of thumb is 400 subjects. However, despite the small samples, results can be considered more robust when applied to specifically defined patient populations with similar demographic parameters. Finally, the number needed to treat (NNT) — ie, the number of patients who must be treated for one patient to benefit — is the single most clinically useful statistic. In the literature reviewed, this statistic provides the likelihood that enzymatic debridement treatment will benefit any individual; thus, this statistic provides a perspective on the reasonableness of a treatment.

**Wound type.** Wound type is clearly identified in each study discussed. This knowledge can empower the wound care specialist to select the most appropriate agent for the particular wound type in the hopes of achieving the same results found in the literature.

**Wound measurement.** Wound measurement before and after invention is available only for six studies. More specifically, wound volume and mean wound size (reported in centimeters) was reported only in one collagenase study. Alvarez et al’s prospective, randomized comparative efficacy trial between collagenase and papain-urea formulations offers objective wound measurements. The remaining investigations offer subjective observations regarding wound measurements and center around the evaluation of papain-formulations. The subjective observation specifically refers to wound closure or healing as an endpoint.

**Therapeutic goals.** Evaluating and applying literature findings to clinical practice is critical to outcome goals. Many outcomes are either disease-oriented evidence that reflects changes in physiologic parameters or patient-oriented evidence such as morbidity, mortality, or cost. The 16 investigations discussed here include clearly defined outcome measures and end-of-study goals that were disease-oriented. Six investigations used duration or time-to-heal as a disease-orientated endpoint, followed by complete healing (five investigations), change in wound environment (two), and wound debridement-wound cleaning or reduction of nonviable tissue (three). All endpoints were evaluated clinically by the investigators or blinded observers.

Although statistical analysis was not provided in many of the 16 investigations, the results may be of clinical value to the wound care specialist. Most important, the viability of the information is directly related to whether the treatment and solution in question are relevant to the wound care specialist’s need.

**Conclusion**

Enzyme products commercially available in the US were reviewed, emphasizing their functional components, mechanisms of action, and patient considerations. A critical literature review of articles relevant to these products discussed search determinants, clinimetric definitions, and search results. A literature search identified 16 enzymatic debridement clinical study publications. However, the majority of investigations had small sample sizes, many did not include a control treatment, and
predicted bias and publication bias were detected in some of the investigations. Most of the evaluations failed to demonstrate grounded statistical significance. This review underscores a need for future studies to be designed to meet the criteria of level A evidence to provide information to the wound care specialist regarding the clinical benefits of enzymatic debridement agents.

References