NECROTIZING SOFT TISSUE INFECTIONS

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Rationale:

Hyperbaric oxygen therapy is a recognized accepted adjunct to surgical debridements, antibiotic therapy and maximal goal-directed critical care therapy for infections of soft tissues resulting in necrosis. A number of clinical scenarios, specific lesions and syndromes have been described over the years, based on the affected tissues and location of infection, the etiologic organism or combination of organisms involved in the infection, and particular host immunologic and vascular risk factors. In all of these clinical situations, there appears to be the common denominator of the development of hypoxia resulting in necrosis.

Hypoxia is known to impair phagocytosis by polymorphonuclear leukocytes.[1] After an infective process is initiated, metabolic products of aerobic and anaerobic metabolism tend to lower the oxidation-reduction potential (E_h), leading to a drop in pH, which creates a milieu for growth of strict and facultative anaerobic organisms. When the blood supply to the skin is affected by involvement within a phlegmon, with edema and necrosis in the deep fascial layers in which they reside, the decreased perfusion pressure and ischemia predispose to rapid progression and advancement of the infectious process within the skin and subcutaneous tissues, exacerbated by the dysfunctioning polymorphonuclear leukocytes. Local hypoxia occurs, with up-regulation of endothelial adherence molecules, resulting in leukocyte adhesion and endothelial cytotoxicity. Leukocytes may become sequestered in vessels, impairing local immunity, and incomplete substrate oxidation results in hydrogen and methane accumulation in the tissues. Tissue necrosis occurs, with purulent discharge and gas production. Quantities of gas within tissues are frequently seen in gas gangrene, crepitant anaerobic necrotizing cellulitis, and necrotizing fasciitis.

Hyperbaric oxygen therapy can reduce the amount of hypoxic leukocyte dysfunction occurring within an area of hypoxia and infection,[1,2,3] and provide oxygenation to otherwise ischemic areas, thus limiting the spread and progression of infection. The diffusion of oxygen dissolved in plasma in the circulation, where it is initially carried in large vessels, proceeds to areas of poorly perfused tissue, from regions of very high O_2 saturation down a gradient to lower oxygen levels in tissue. Integrin inhibition decreases leukocyte adherence, reducing systemic toxicity.[5]

In cases where the antibiotic being used requires oxygen for transport across cell walls, hyperbaric oxygen therapy can act to enhance antibiotic penetration into target bacteria. Enhancement of the post-antibiotic effect by hyperbaric oxygen has been demonstrated for aminoglycosides and Pseudomonas.[6]

Clinical classification of the necrotizing infections of soft tissues is easiest early in the course of infection, when anatomic levels of involvement of skin, superficial or deep fascia, and muscle involvement can be assessed either during exploration, on punch biopsy, or by radiologic investigation. However, as infection progresses, distinction between some of the clinical entities may become blurred as full thickness necrosis extends into muscle late, after having extended

through skin, fat, fascia, and into muscle via direct extension of infection. At presentation, it may be difficult to differentiate these necrotizing soft tissue infections one from another, or from Clostridial myositis and myonecrosis, until either Gram stain or cultures are available. Considering their historical differences and evolution, it remains useful to examine the separate categories of infection separately in order to anticipate pathways of extension of infection, anticipate complications, and identify when adjunct hyperbaric oxygen therapy should be considered.

Clinical Entities: Necrotizing Fasciitis

Introduction: Necrotizing fasciitis is an acute, potentially fatal infection of the superficial and deep fascia of the skin and soft tissues, which progresses to ischemic dermal necrosis after involvement of the dermal blood vessels which traverse the fascial layers. The popular media refer to this entity as infection with "Flesh-eating bacteria."

Etiology

Necrotizing fasciitis was initially described and named "hemolytic streptococcal gangrene" by Meleney in 1924.[7] He described an illness characterized by gangrene of subcutaneous tissues, followed by rapid necrosis of the overlying skin from involvement of the blood vessels supplying the skin, which are found in the affected fascial layers. All his patients grew hemolytic streptococci on cultures, and the patients were all seriously ill. Surgical extirpation appeared to be the therapeutic approach. Reference to this entity as necrotizing fasciitis appears around the time of the report by Wilson.[8]

The characteristic level of infection is at the deep fascia. Because infection with necrosis is noted to spread along fascial planes deep to the skin, it is not an uncommon event for there to be minimal skin signs early on. Pain out of proportion to findings could be an early tip off to the presence of deep fascial infection. Since blood vessels supplying overlying skin travel thru fascia, it is the involvement of these vessels by infection that leads to rapid progression to dermal necrosis. Microbiologically, groups A, C, or G beta-hemolytic streptococci can be isolated from tissue specimens in 50 to 90% of case series, with one or two more organisms often also accompanying the streptococci in up to half the cases. The occurrence of *Staphylococcus aureus* plus anaerobic streptococci is also known as Meleney's synergistic gangrene. Commonly isolated organisms include Enterobacteriaceae, Enterococci, Bacteroides species, Peptococcus species. Candida species have also been reported.[9] Necrotizing fasciitis is also reported to be caused by community-acquired strains of methicillin-resistant *Staphylococcus aureus* (CA-MRSA) alone.[10] In many cases, infection is polymicrobial, with Enterobacteriaceae and anaerobes frequently isolated.

Risk factors

The most common risk factors associated with necrotizing fasciitis are traumatic breaks in the skin, most commonly lacerations, insect bites, burns, deep abrasions, puncture wounds, or following surgery, particularly those involving bowel perforations. Diabetes appears to be a strong risk factor, as are obesity, alcoholism, smoking, and intravenous drug abuse. Reports of

necrotizing fasciitis as a result of infection of otherwise typical lesions of chickenpox have been published.[11] An association with the use of non-steroidal anti-inflammatory agents has also been suggested.[12,13] NSAIDs are cyclo-oxygenase inhibitors and may have an adverse effect on neutrophil killing and cell-mediated immunity. NSAIDs are reported to inhibit monocyte superoxide production.[14]

Most common sites of occurrence of necrotizing fasciitis are the lower extremities, while an increased incidence in the upper extremities is seen in the parenteral drug abuse population. However, any location of the body can be affected, including the abdominal wall of neonates, in association with omphalitis.[15] Involvement of the scrotum and perineum in the male is known as Fournier's Gangrene, which is essentially necrotizing fasciitis of the superficial perineal fascia, also known as Colles' fascia; which can spread infection to the penis and scrotum via Buck's fascia or Dartos' fascia; or Scarpa's fascia, which connects to, and can spread infection to, the abdominal wall. Perianal or perirectal infection may also spread into these areas, and undrained or inadequately drained perirectal abscesses are often cited as a source of Fournier's Gangrene. Perineal necrotizing fasciitis can also occur in the female. Diabetes mellitus remains a strong risk factor in this particular form of necrotizing fasciitis as well. Fournier's Gangrene is more likely to have multiple mixed organisms cultured, particularly Enterobacteriaceae, Group D streptococci, and anaerobic organisms, such as *Bacteroides fragilis*.

Clinical Presentation

The patient with necrotizing fasciitis will typically present with an acute combination of pain and swelling, which may or may not be accompanied by fever and chills. There may already be a focus of cellulitis apparent, but in some instances early on, there may be very few skin changes. In some patients, there may be pain out of proportion to the skin findings, which may not be unexpected considering that the initial level of infection is the fascia, not necessarily the skin. In others, manifestations of a large phlegmon may be quite obvious, although at times the area of infection may have been assumed to be cellulitis and not a more serious form of infection. Pain may proceed to numbness, as a result of compression of nerves which also pass through the fascia. With time however, the infection will rapidly proceed to cause areas of blistering and bullae formation. Hints of darkening of the skin may appear as perfusion decreases, until obvious areas of dermal ischemia appear, making the skin appear dusky, grayish or frankly black. Upon exploration of the process, a clinical diagnosis can be confirmed at the time of biopsy or debridement, when the fascia is grossly observed by the surgeon to be necrotic, and will give way easily to a probing finger or surgical clamp, giving the sensation of "thunking" of the skin against the underlying muscle layers, instead of remaining tight and crisply defined. It has been suggested that limbs of patients with necrotizing fasciitis, as opposed to those with cellulitis only, may be observed to have markedly reduced tissue oxygen saturations as measured by near - infra-red spectroscopy throughout the involved site, with oxygen saturations in the 52% ±18% range, compared to control measurements of 86% ±11% in uninvolved sites.[16]

In the neonate, necrotizing fasciitis of the abdominal wall can be seen as a complication of omphalitis in 10 to 16% of cases,[17] and appears to carry over a 50% mortality rate even when treated with aggressive debridement of involved skin, subcutaneous tissue and fascia.[18]

A number of diagnostic observations have been made to enable confirmation of the diagnosis of necrotizing fasciitis. Frozen section soft-tissue biopsy early in the evolution of a suspect lesion may provide definitive diagnosis.[19] CT scan findings are also revealing. Asymmetrical fascial thickening that was at least twice the contralateral side and associated with fat stranding was seen in 80% of 20 patients with necrotizing fasciitis. Gas tracking along fascial planes was seen in 55%, characteristically did not involve muscle and was not associated with abscess formation.[20] The authors note that the areas of black, gangrenous skin were far smaller than the widespread infection in the underlying fascial planes. Also of note was that 7 of the 20 patients had associated deep space abscesses requiring immediate surgical drainage, which demonstrates the need for CT studies to assess extent of disease, particularly in patients who do not appear to be responding to therapy.

Magnetic resonance imaging (MRI) also demonstrates the extent of affected tissue well, is able to differentiate fluid and gas through differential signal intensities, and is useful in differentiating cellulitis from necrotizing fasciitis, after injection of gadolinium contrast. But in a study of 15 patients, MRI overestimated the extent of deep fascial involvement in one patient who only had cellulitis, following IM injections which showed up on MRI as thickening of both superficial and deep fascia of the deltoid muscle.[21]

Cultures of deep tissue at the time of debridement for aerobes, anaerobes and fungi, are imperative as up to 75% of patients in some series' have demonstrated polymicrobial etiologies. Fungal cultures are particularly important in the immunocompromised, diabetic and cancer populations and in patients who have not responded to standard anti-bacterial antibiotics.

Amputation rates of 26%[22] up to 50%[23] are reported in cases of necrotizing fasciitis of the extremities, without hyperbaric therapy. Mortality in reported series range from 16.9% up to 66% without the use of hyperbaric oxygen. Mortality is often associated with delayed diagnosis, underlying immunocompromise, and underlying heart disease, degree of leukocytosis, septic shock and severe underlying metabolic abnormalities.

In the neonate, necrotizing fasciitis is reported as a complication of omphalitis, balanitis, mastitis, postoperative complication, and fetal monitoring.[24] 4 of 6 cases found in a literature review who received hyperbaric oxygen therapy survived, while the overall mortality rate was 39/66 (59%). In a group of neonatal omphalitis patients with abdominal wall necrotizing fasciitis reported from Children's Hospital in Los Angeles, 7 out of 8 cases died, for a mortality rate of 87%[25] without hyperbaric oxygen therapy. In a series of 32 cases of omphalitis from Seattle over a 10-year period, 7 developed necrotizing fasciitis, and 5 of the 7 died. The 2 patients who did survive, out of the 4 who had hyperbaric oxygen treatments, were noted to have resolved their systemic sepsis more rapidly, and had healthier granulation tissue on the perimeter of the debridement. Neither survivor treated with hyperbaric oxygen required any further debridements before their wounds were closed.[26]

Gozal et al[27] treated necrotizing fasciitis patients with combined antibiotics, radical surgery and hyperbaric oxygen, and reduced the historic mortality rate from 38% to 12.5%. Of 29 patients reported retrospectively by Riseman et al,[28] 12 were treated by surgical debridement and antibiotics only, and 17 received hyperbaric oxygen treatments in addition. Both groups had

similar parameters of age, race, sex, wound bacteriology and antimicrobial therapy. Body surface area was also similar. However, perineal involvement (53% vs. 12 %) and septic shock (29% vs. 8%) were more common in the hyperbaric group, yet the overall mortality was significantly lower at 23%, versus 66% in the non-hyperbaric oxygen treated group. Additionally, only 1.2 debridements per patient in the hyperbaric treatment group were performed, vs. 3.3 debridements per patient in the surgery plus antibiotics-only group.

Differential Diagnosis

Clearly a goal when making the diagnosis of necrotizing fasciitis is to make it as early as possible so as to be able to start appropriate treatments and avoid rapid spreading and the onset of sepsis. Time is tissue. The main differential diagnoses includes standard cellulitis, which may be a precursor of necrotizing fasciitis in some cases; and erysipelas, with its erythematous well-delineated border. Additional entities which should be considered include Clostridial myositis and myonecrosis; non-Clostridial myositis and myonecrosis; toxic shock syndrome, which may accompany necrotizing fasciitis; Zygomycotic gangrenous cellulitis; mixed aerobic/anaerobic necrotizing cellulitis; toxic epidermal necrolysis (TEN), also known as Lyell's Disease, usually due to exposure to particular medications; and Staphylococcal Scalded Skin Syndrome, also known as Ritter's Disease, due to exfoliative toxins produced by Staphylococci, with the latter two entities being most common in neonates and children under 5 years of age. In the neonate with omphalitis, violaceous discoloration of the skin appears to be a strong marker for the emergence of necrotizing fasciitis. *Vibrio vulnificans* infections cause blistering infection quite commonly, and are seen in patients who have either been swimming in waters, along the Gulf of Mexico, or have been eating shellfish from that area. *Aeromonas* infections also occur following open wounds acquired in sea water. Cutaneous anthrax may present with a blackened central area and surrounding edema.

Clinical Management

Numerous studies have continued to demonstrate the beneficial effect of hyperbaric oxygen therapy in the management of necrotizing fasciitis. Wilkinson and Doolette[29] reported a 5- year retrospective cohort Australian study of 44 patients with necrotizing soft tissue infection, between 1994 and 1999, looking at the primary outcome of survival to hospital discharge, and secondary outcomes of limb salvage and long-term survival after hospital discharge. Logistic regression analysis determined the strongest association with survival was the intervention of hyperbaric therapy (p=.02). Hyperbaric oxygen therapy increased survival with an odds ratio of 8.9 (95% confidence interval, 1.3-58.0) and a number of 3 needed to treat to benefit. Hyperbaric oxygen therapy also reduced the incidence of amputation (p=.05) and improved long-term outcome (p=.002). In the series by Escobar et al, there were no further amputations beyond those already done prior to transfer, once hyperbaric oxygen therapy was initiated in their series of 42 patients.[30] The negative study by Brown at al[31] which purports to be a multi-center retrospective review of treatment at 3 facilities over 12 years, of 54 patients, had numerous discrepancies in the demographics of their two groups. Half of the hyperbaric-oxygen-treated group of 30 patients, all from one institution, were noted to have Clostridial infections, while the non-hyperbaric treated group had only 4 of 24 patients (17%) with Clostridial infection. 6 of the

30 in the hyperbaric group are noted to have the diagnosis of Clostridial myositis and myonecrosis, whereas only one of the non-hyperbaric oxygen treated patients were so diagnosed. Hence this clearly shows the same diseases were not being compared in this study. Additionally, as is pointed out in a subsequent letter to the editor,[32] 80% of the patients received 4 or fewer treatments, the remaining 20% received between 5 and 7 treatments, and the timing of these treatments is not specified. If the guideline of treating three times in the first 24 hours were followed, and then twice per day until the patient is stable and shows no relapse of toxicity between treatments, the gas gangrene patients in this study were treated for less than a day and a half, which is a shorter period of time than most other studies, and the others were treated for around 2 days. In the Wilkinson study, patients received a median of 8 treatments, which is more than that received by the patient with the greatest number of treatments in Brown et al. The authors state that the mortality difference between the two groups (9/30, or 30% of the hyperbaric group, versus 10/24, or 42% in the non-hyperbaric group) was not statistically significant. Thus the Brown et al study should not be used as an argument that the use of hyperbaric oxygen for truncal necrotizing fasciitis is "controversial," because these mortality statistics are not comparable, with a different mix of diagnoses in the two, compounded by the fact that the numbers themselves are small, resulting in a study that had insufficient power to demonstrate a statistically significant result.. Furthermore the study does not add to the literature of necrotizing fasciitis involving the limbs and other non-truncal sites.

Fortunately, Fournier's Gangrene cases in the literature are usually studied and reported as a distinct group. Hollabaugh et al[33] reported a retrospective series of 26 cases from the University of Tennessee's five hospitals. Of the 15 patients with identifiable sources for their infections, 8 had urethral disease or trauma, 5 had colorectal disease, and 2 had penile prostheses. All patients were managed with prompt surgical debridement and broad-spectrum antibiotics. Procedures performed included urinary diversion, fecal diversion, and multiple debridements. Fourteen of the twenty-six were additionally treated with hyperbaric oxygen. The group treated with hyperbaric oxygen had a mortality rate of 7%, versus 42% in the group not receiving hyperbaric oxygen ($p=.04$), with a combined overall mortality rate of 23%. The one patient who died while receiving hyperbaric oxygen therapy had been progressing well without evidence of ongoing infection, but suffered an acute MI not thought to be related to the underlying disease process. In the non-hyperbaric group, deaths were usually attributed to ongoing or fulminant sepsis. Relative risk for survival was 11 times greater in the group receiving hyperbaric oxygen therapy. This study did not show a decrease in the number of debridements by HBO_2 therapy, but was confounded due to the larger number of patients who died and thus were not able to get further debridements. Delay to treatment was not a factor in the different groups.

Additional series include that of the Genoa, Italy group[34] who treated 11 patients without any deaths, and all delayed corrective procedures healed without infectious complications. Another 33 patients were reported in a series from Turku, Finland.[35] These patients were treated at 2.5 atm abs, in conjunction with antibiotics and surgery. 3 patients died, for a mortality rate of 9%. Hyperbaric oxygenation was observed to reduce systemic toxicity, prevent extension of the necrotizing process, and increased demarcation, improving overall outcomes. 2 of the 3 patients who died were moribund upon arrival to their facility. Management included diverting colostomies for those patients with a perirectal or perineal source, and orchiectomy, although sometimes reported in all series, is not routinely done since the blood supply to the testes is from

the spermatic vessels which do not perfuse the scrotum and penis. Suprapubic cystostomy was indicated and performed when the source of the infection was genitourinary.

Due to the difficulty in making direct comparisons of clinical series', a Fournier's Gangrene Severity Index Score was developed[36] in order to assess a number of variables rather than the presence of the disease itself. The score uses degrees of deviation from normal of physiologic variables to generate a score that correlates with patient mortality. It is clear that the amount of disease, related by some to body surface area of involvement, may be a significant variable. The Duke University analysis of 50 consecutive patients seen at their institution over a 15 year period had a 20% overall mortality.[37] Three statistically significant predictors of outcome were identified when examined using univariate analysis: extent of infection, depth of the necrotizing infection, and treatment with hyperbaric oxygen. However the same data using multivariate regression analysis identified only the extent of the infection as the only statistically significant independent predictor of outcome in the presence of other co-variables. Patients with disease involving a body surface area of 3.0% or less all survived. The numbers of patients with disease extent greater than 3%, where hyperbaric oxygen would thus be expected to play a role, became smaller, and with small numbers of patients, the power of the study to demonstrate a significant response was not present. Using multivariate analysis, the p value for statistical significance for hyperbaric oxygen treatments was equal to .06.

With such strong case series evidence of reductions in morbidity and mortality for necrotizing fasciitis and the subset of Fournier's Gangrene, it is difficult to envision ever seeing a controlled, double-blinded study of hyperbaric oxygen therapy.

Patient Selection Criteria

Patients who are candidates for hyperbaric oxygen treatments will have had the diagnosis of a necrotizing soft tissue infection made. Strongest consideration should be given to patients who are compromised hosts, as they are likely to do the worst with their infection. Significant organ failure is not a contra-indication to treatment, as long as the treating chamber facility is properly staffed to manage the specific complications of the individual patient, particularly those on ventilators and pressors.

Clinical Management

The recommended hyperbaric oxygen treatment protocol for necrotizing fasciitis includes initiating therapy at 2.0 to 2.5 atm abs pressure for 90 minutes of oxygen given twice a day for the first few days, until there appears to be no further extension of necrosis in previously debrided areas, and infection is "controlled."[38] If there is doubt of the diagnosis and Clostridial myositis and myonecrosis are still in the differential diagnosis, higher pressure treatments of 2.8 to 3.0 atm abs should be used, using the gas gangrene protocol of 3 treatments in the first 24 hours. Some hyperbaric specialists may switch to once per day treatments once the patients appear stabilized to be sure that the process does not relapse, prior to stopping. Hyperbaric oxygen therapy is an adjunct to, and does not substitute for, standard wound care, debridement of necrotic tissue, drainage of fluid collections and abscesses, use of antibiotics directed at the

expected range of organisms, use of intravenous gamma globulin, particularly if the necrotizing soft tissue infection is associated with Group A hemolytic streptococcal infection and toxic shock syndrome,[39] and goal-directed management of sepsis.

OTHER NECROTIZING BACTERIAL INFECTIONS

Non-Clostridial Myonecrosis

This is a particularly aggressive soft tissue infection, which clinically acts much like the Clostridial myositis syndrome, with widespread involvement of muscle and fascia. It has also been called "Synergistic Necrotizing Cellulitis." [40] It is differentiated from necrotizing fasciitis by the muscle involvement, although infection from necrotizing fasciitis, if left to progress, will ultimately spread into muscle, and may be indistinguishable from Non-Clostridial myositis at that point. Organisms described to be involved include the anaerobic *Peptococcus* species, *Peptostreptococcus* species, and *Bacteroides* species, often mixed with aerobic members of the *Enterobacteriaceae*.[41] Clinically, the patient will present with exquisite local tenderness, minimal skin changes, and drainage of "dishwater" pus from skin surface ulcerations, which become enveloped in blue-gray gangrene. Most patients are quite ill systemically. Half of the patients are bacteremic. Gas can also be seen. This is often described as the entity when Fournier's Gangrene extends onto the abdominal wall and pelvis, involving muscle and fascia alike. Treatment remains surgical debridement. Since there is a very frequent component of anaerobic organisms in this entity, it would seem reasonable to use the same rationale as for treatment of necrotizing fasciitis, and a similar treatment protocol.

Crepitant Anaerobic Cellulitis

This category encompasses both Clostridial and non-Clostridial skin infection. There is abundant tissue gas, but no fascial or muscle involvement. When Clostridial species are present in this situation, the conditions are not conducive to toxin formation, and the patient will lack marked systemic toxicity. It is most commonly reported after local trauma to the lower extremities in patients with vascular insufficiency. Organisms reported include *Clostridium* species, *Peptococcus* species, *Peptostreptococcus* species, *Bacteroides* species, and *Enterobacteriaceae*. Gas formation causes the typical "crepitance" palpable within the skin. Antibiotics and surgical therapy in normal hosts is usually adequate therapy. Hyperbaric oxygen therapy should be considered in compromised hosts and in those failing to respond. Mortality rate is given at around 10%.

Progressive Bacterial Gangrene

This is a subacute process, characterized by slowly progressive dermal ulceration, usually found on the abdominal wall or thorax. It was first described by Cullen in a patient after drainage of an appendiceal abscess.[42] It does not extend to deep fascia. It usually develops at a surgical site, such as a colostomy or ileostomy site. The area around the wound becomes erythematous, swollen and tender, with progression to induration. A central purple area develops, and proceeds to slough off as the lesion enlarges and develops a granulation area centrally, surrounded by a gangrenous margin. The pathology is said to be related to progressively expanding infection

created by the synergism between aerophilic and anaerobic/microaerophilic bacteria. It is thought to be similar to, or be identical to, Meleney's ulcer, which has as its hallmark a progressive, slowly extending rim of necrosis, which may tunnel subcutaneously and spread in an occult fashion. It is also seen following lymph node surgery in the neck, axilla or groin. Hyperbaric oxygen therapy has been shown to lead to improvement when other standard therapies have failed.[43]

ZYGOMYCOTIC GANGRENOUS CELLULITIS

Rationale

Introduction: In the immunocompromised population, infection with opportunistic organisms is a not uncommon occurrence. Opportunistic organisms typically do not cause disease in normal host patients, but due to particular deficits in the immune response of various categories, these otherwise unusual organisms become common findings in the abnormal host population. Until now, the discussion has centered on bacterial and bacterial toxin-induced diseases, but fungal organisms may also become significant pathogens in that population of patients. A significant virulence factor of these organisms is their characteristic invasion of blood vessels, causing ischemia, hypoxia, and progressive necrosis of tissue, thus creating a niche which would physiologically appear to be amenable to alteration through the use of hyperbaric oxygen therapy. This form of infection is certainly considered one of the necrotizing soft tissue infections.

Etiology

Zygomycosis is the name given to the group of fungal infections caused by pathogenic molds belonging to the class Zygomyces, in the phylum Zygomycota. The term "Phycomycosis" has also been used, but is less commonly used today. Zygomyces is further divided into two orders, *Mucorales* and *Entomophthorales*. The Mucorales usually cause infections that are acute in onset, aggressive, rapidly progressive, and angio-invasive. These infections are commonly called Mucormycoses. In the family Mucoraceae within the order Mucorales are organisms of the genera *Absidia, Apophysomyces, M ucor, Rhizomucor,* and *Rhizopus*. Additional less common families include Cunninghamellaceae with organisms of the single genus *Cunninghamella*, and Saksenaceae, with the single genus *Saksenaea*, and others.[44] Organisms in the order Entomophthorales are *Conidiobolus coronatus* and *Basidiobolus ranarum*. These produce a group of infections that tend to be more indolent, but clearly pathologic and chronically progressive. They typically do not invade blood vessels, although some recent reports do suggest that this may occur at times.

Risk factors

The recognized risk factors for Zygomycoses are numerous. The leading risk factor appears to be diabetes mellitus, particularly in the setting of keto-acidosis or uncontrolled hyperglycemia. It is reported that 70% of cases of rhino-cerebral Zygomycosis occur in the setting of keto-acidosis.[45] The acidotic environment is said to be ideal for fungal growth, while white blood cell activity is inhibited in the hyperglycemic environment.[46,47,48] It has been shown that

acidosis disrupts the inhibitory activity of sera against fungal growth by interrupting the capacity of transferrin to bind iron, which would normally keep it from being available to the fungal species.[49] Another group of patients at risk are those with iron overload syndromes who are at risk for more significant infections due to the presence of higher levels of iron, a growth factor for most bacteria and fungi capable of synthesizing endogenous metal chelators, or siderophores; or in patients on metal chelators, such as dialysis patients receiving deferoxamine[50,51] for removal of aluminum. Since deferoxamine is normally cleared by the kidney, levels of the drug remain high in the dialysis population, prolonging the time that iron bound to it can be utilized by the fungi. Other susceptible patients are those with underlying malignancies, especially leukemias; patients with neutropenia; solid organ and bone marrow transplant patients, and patients who are actively or passively immunosuppressed. Patients who have been on broad-spectrum antibiotics may have fungal overgrowth, which is also a risk factor. The organisms are ubiquitous fungi, and commonly inhabit decaying matter such as common garden soil. Introduction of infection is often related to antecedent trauma.[52] A history of exposure to organisms through farm accidents or trauma in the garden would not be unusual. GI involvement is associated with extreme malnutrition, and is though related to oral ingestion of spores of the organisms. Around 5% of patients appear to have no risk factors whatsoever.

Clinical presentations

The most common manifestations of Zygomycosis are sinusitis, rhino-cerebral infection, soft tissue infection, pneumonia, gastrointestinal involvement, and disseminated infection. In the sinusitis and rhino-cerebral forms of the infection, initial symptoms would be similar to routine sinusitis, with sinus pain, congestion, and drainage. The infection then accelerates, extending into adjacent structures and tissues, with development of erythema, progressing to violaceous or dusky to frankly black tissue in the nares, turbinates, palate or orbit. The organisms appear to have a predilection for invasion of arteries, lymphatics and nerves. Invasion of vascular structures leads to a fibrin reaction and development of a Mucor thrombus within vessels, which leads to infarction. The infarcted tissue becomes acidotic and permissive for even further fungal ingrowth and proliferation. Lack of perfusion prevents antibiotic penetration into affected tissues. Extension into adjacent periorbital and orbital structures is often found even early on. Clinical manifestations can include periorbital edema, tearing and proptosis. Involvement of the optic nerve will be marked by blurring, followed by loss, of vision. Abnormalities of eye movement may occur as markers of cranial nerve involvement. Extension can also move inferiorly into the hard palate via the maxillary sinuses; black, necrotic ulcers may be found on the palate, and the nasal turbinates may appear black and necrotic. Infection may extend into the cranial vault, either via the ethmoid sinus and through the cribriform plate, or through the orbital apex into the area of the cavernous sinus, producing the Orbital Apex Syndrome, consisting of ophthalmoplegia and 5th cranial nerve involvement, progressing to cavernous sinus thrombosis, and thrombosis of the internal carotid artery, resulting in major hemispheric stroke and altered consciousness. Due to the propensity for angio-invasion, fungemia can occur, disseminating the infection systemically. Rhino-cerebral mucormycosis has a very high mortality rate. Standard treatment consists of the antifungal antibiotic Amphotericin B lipid complex or liposomal Amphotericin B, in a dose of 5 mg/kg daily and surgical debridement when indicated. Survivors have usually had earlier diagnosis and surgical debridements.

Pulmonary involvement is the second most common type of Zygomycosis overall, seen particularly in patients with leukemia and lymphoma.[53] Isolated solitary nodular lesions, lobar involvement, cavitary lesions and disseminated lesions have all been reported.[54] Erosion of the fungus into mediastinal structures, particularly the pulmonary artery, with massive hemoptysis, is a fatal occurrence. Wedged infarctions of the lung may be seen, as a manifestation of thrombosed pulmonary vessels, from angio-invasion.[55]

One of the manifestations of cutaneous infection includes a rapidly progressive, ascending, necrotizing infection consistent with necrotizing fasciitis, which can involve an extremity or the torso. Aerial hyphae can sometimes be grossly visualized in wounds infected with Zygomycosis organisms, as a loose, whitish cottony exudate covering the surface of open wounds. Risk factors for the development of cutaneous and subcutaneous involvement include various types of breakdown of the skin barrier, including puncture wounds, other trauma, and burn wounds. Mortality rates of 30% to 70% are reported in necrotizing fasciitis with these organisms, depending on the underlying condition associated with the infection. Since diabetic ketoacidosis is a treatable condition, reversal of the acidosis affords an opportunity for the host response to reconstitute, and thus may have a decreased mortality compared to the patients with non-reversible conditions.

The GI syndrome is characterized by abdominal pain and distention, associated with nausea and vomiting. Fever and hematochezia may occur. Stomach, ileum and colon are most commonly affected. Most such diagnoses are made post-mortem, but, if suspected, may require laparotomy to manage the bowel infarctions that may occur.[56]

Differential diagnosis

Upon initial presentation, rhino-cerebral mucormycosis may be misidentified as the more common routine bacterial sinusitis due to usual Gram positive or anaerobic organisms, although there should not be any necrotic lesions in those cases. However, once evidence of necrosis is apparent, or in the proper clinical settings, there should be no hesitation in ordering a biopsy, looking for the various fungal forms, which are quite characteristic wide, non-septate hyphae branching off at right angles; and signs of angio-invasive processes should be sought. Affected tissue usually has neutrophilic infiltrates and inflammatory vasculitis is seen, involving both arteries and veins. Cultures for routine aerobic, anaerobic and fungal organisms should always be sent. Cavernous sinus thrombosis can occur as an extension of suppurative, usually Staphylococcal, facial cellulitis or abscess, but there would not be the typical lesions in the nose or sinuses. Radiological studies, such as plain films or CT scans, may show more extensive bone necrosis than was anticipated. Orbital cellulitis and bacterial osteomyelitis of the frontal bone or orbit are other entities which may clinically resemble this form of Zygomycosis.

Lung involvement may be non-specific, and can look like other cases of atelectasis, pneumonia, granulomatous disease or, particularly in patients with cancer, infection due to *Aspergillus* species. Use of radiologic studies may hasten the diagnosis. In a retrospective analysis of CT findings in 16 cases of pulmonary Zygomycosis vs. 29 cases of invasive pulmonary aspergillosis at the University of Texas M.D. Anderson Cancer Center,[57] logistic regression analysis of clinical characteristics demonstrated that a) concomitant sinusitis and b) voriconazole

prophylaxis were significantly associated with pulmonary Zygomycosis; CT scan findings of multiple (≥10) nodules, and pleural effusion were both independent predictors of pulmonary Zygomycosis, suggesting potential clues in differentiating the two types of infections. Pulmonary mucormycosis can also be confused with standard pulmonary embolism. Gastrointestinal disease must be differentiated from other bowel infections, perforation and Staphylococcal Necrotizing Enterocolitis, seen in infants.

Rationale for use of hyperbaric oxygen therapy

From a physiological viewpoint, mechanistic steps are only now being discovered to explain the virulence and invasiveness of the filamentous fungi in causing disease. Each of these mechanisms, as discovered, would well be worth testing in the presence of hyperbaric oxygen to assess potential roles for hyperbaric oxygen therapy. Filamentous fungi are aerobic and thus it is not expected that there would be a direct effect on fungi under clinical hyperbaric conditions.
Hyperbaric oxygen therapy in the setting of Zygomycosis could be beneficial in a number of ways. The angio-invasive character of these infections creates areas of hypoxia, ischemia and subsequent necrosis, which will directly affect neutrophilic killing of organisms, as phagocytosis becomes inefficient. Areas of tissue that are ischemic due to partial loss of perfusion can be made normoxic during hyperbaric therapy, and restore immune mechanisms that have become dysfunctional due to hypoxia.

The neutrophil has a significant role in defending against filamentous fungi, despite the larger size of the hyphae. Engulfment by neutrophils and damage to hyphae is correlated with response to infection. Both mononuclear and polymorphonuclear white cells of normal hosts kill *Rhizopus* by generation of oxidative metabolites and cationic peptide defensins.[58,59,60] Comparison of antifungal function of human polymorphonuclear leukocytes against hyphae of *Rhizopus oryzae* and *Rhizopus microsporus*, the most frequently isolated zygomycetes, with that of *Absidia corymbifera* have shown that oxidative burst responses by PMNs, and PML-induced hyphal damage, were significantly lower in response to the *Rhizopus* species than to the *Absidia* species, and that hyphal damage increased when PMLs were incubated with Interferon-gamma and granulocyte-macrophage colony stimulating factor (GM-CSF).[61] Mouse bronchoalveolar macrophages prevent germination of spores in vitro and in vivo in a murine model, and this ability is blocked by corticosteroid therapy. Correction of hypoxia for such critical cells should enhance oxidative killing of fungi. The significant hallmark of Zygomycoses is their ability to invade blood vessels, causing blood vessel inflammation, thrombosis and tissue necrosis in many different tissues, and subsequent hematogenous dissemination to other organs. Penetration of endothelial cells lining blood vessels must be a key step in the pathophysiology of Zygomycosis. Studies examining these steps are crucial in defining additional steps to treat infection, by blocking fungal dissemination. It has been demonstrated that *Rhizopus oryzae* spores adhere to subendothelial matrix proteins better than hyphae, but spores and hyphae adhere equivalently to human umbilical vein endothelial cells.[62] Phagocytosis of *Rhizopus oryzae* by endothelial cells was also shown to damage the endothelial cells, raising the question of whether such steps could be related to subsequent thromboses. Hyperbaric oxygen research has not begun to delve into these neutrophil and fungal/endothelial interactions, but is sorely needed.

Much of the surgery required to manage the necrotizing aspects of infection involving sinuses, orbit and skull is quite deforming, and the addition of hyperbaric oxygen to wound management would facilitate generation of granulation tissue, epithelialization, and bone healing. Additionally, there are other non-specific mechanisms that are still being worked out for several forms of sepsis, which appear to be positively affected by hyperbaric oxygen.[63,64]

Standard therapy involves the use of antifungal antibiotics, and definitive debridement of necrotic tissue. Hyperbaric oxygen clinical studies to date have generally been either isolated case reports, or retrospective case series and literature reviews. John et al[65] reported such a literature review of 28 published cases that had received hyperbaric oxygen treatments. Among the Mucorales isolates, there were 11 cases of *Rhizopus* species, followed by 3 cases of *Apophysomyces* species, and 2 cases each of *Mucor* and *Absidia*. Three isolates from Entomophthoramycoses were *Conidiobolus* species. Risk factors in these patients were a spectrum of the typically seen range, with 17/28 (61%) being diabetics, 10 of whom had ketoacidosis; 5 patients (18%) developed their infections after trauma, one patient was on systemic steroids, 3 (11%) had hematological malignancies or bone marrow transplants, and 3 (11%) had no known risk factors for Zygomycosis. Overall survival rate was 86%, which encompassed a 94% survival rate in diabetic patients, but only a 33% survival rate in patients with hematological malignancies or bone marrow tranplants. All patients except for two had also received Amphotericin B. Despite the range of cases, all groups were small, and there were no case controls to which to compare the case responses.

In a large series of all cases of Zygomycosis found in the literature since 1885, 929 cases of Zygomycosis were reported and analyzed by Roden et al.[66] Survival rates were reported by type of treatment received. 44 patients were identified as having received hyperbaric oxygen therapy, and in that group, 64% of patients survived. Other treatments identified and survival rates were: Amphotericin B deoxycholate recipients: 324 survivors of 532 patients (61%); Amphotericin B lipid formulation- 80 survivors of 116 patients (69%); Itraconazole, ketoconazole or posaconazole- 10 survivors of 15 patients (67%); No antifungal therapy at all- 59 survivors of 333 patients (18%); Surgery alone- 51 survivors out of 90 patients (57%); Surgery plus antifungal therapy- 328 survivors out of 470 cases (70%); Granulocyte colony-stimulating factor-15/18 (83%); Granulocyte transfusion- 2 survivors out of 7 cases (29%); in patients who received no therapy at all, there were 8 survivors out of 241 cases (3%). Major difficulties arise with these data, particularly since these studies usually do not differentiate between intent to treat studies or salvage therapy when standard treatment appears to be failing, whether the cases related to use of antibiotics, surgery, or hyperbaric oxygen therapy. This is an observed difficulty in interpreting large numbers of individual case reports and series.[67] The case reported by Bentur et al of mucormycosis of the 4th finger of the hand, in a diabetic with ketoacidosis, is such a case,[68] where hyperbaric oxygen therapy is begun only after other modalities, including Amphotericin B, amputation of the affected finger, followed by wide debridement of the hand, and fasciotomy of the forearm have been tried and the disease continued to progress. After receiving 29 hyperbaric treatments, the infection appears improved, and the patient went on to heal her wounds. Similarly, the case of an *Entomophthorales* infection in the medial orbit of an 18 month old is another example of hyperbaric oxygen therapy used as salvage therapy[69] in conjunction with radical surgery, when the organism was found to be resistant to all available anti-fungal antibiotics. Thus any future database of cases of

Zygomycoses treated with hyperbaric oxygen therapy should document classification of cases by whether hyperbaric oxygen was used as an early adjunct, at the time of initial institution of therapy, or as "rescue," or "salvage" therapy. In addition, hyperbaric oxygen would normally be considered an adjunct to use of antibiotics and indicated surgery, and such sub-group analysis was not done in the Roden report. It is unfair to compare the results if hyperbaric oxygen therapy were started late in the treatment course, as salvage therapy, when an initial course of antibiotics and surgical debridement have been determined to have failed, and infection is progressing and considered to be refractory, as opposed to surgery and antibiotics, started early on, without hyperbaric oxygen. Appropriate comparisons can be made when hyperbaric therapy is added as an adjunct to the initial management of surgery and antibiotics. A strong argument for controlling for such variables in different studies is well-advised, and is comparable to the discussions in the medical literature related to other salvage interventions, such as a new antibiotic, where the answers to the questions of how much of the treatment effect is attributable to the commencement of therapy, and how much is attributable to the natural history of partially-treated disease, can rarely be separated out.[70] In the setting of a rare, relatively unusual infection, it is a given that randomized studies would be unrealistic, and those authors recommend that carefully selected, matched, contemporaneous control subjects are likely to be the most useful alternative. Although these comments were made in reference to use of newer antifungal antibiotics, the same observations would apply to the analysis of hyperbaric oxygen therapy.

Treatment

Antibiotic treatment should be commenced with an Amphotericin B preparation. The fungus is relatively refractory to standard medical therapy, thus maximally-tolerated doses of Amphotericin B deoxycholate should be used, usually 1.0 to 1.5 mg/kg/day. Lipid complex forms of Amphotericin B doses are better tolerated, and doses are higher. The dose of Amphotericin B lipid complex (Abelcet) and liposomal Amphotericin B (AmBisome) is 5 mg/kg/day. It has been observed that the use of voriconazole as fungal prophylaxis in the hematopoietic stem call transplant population is a risk factor for developing Zygomycosis[71] and should be avoided. Other currently available azoles, such as ketoconazole, itraconazole, fluconazole or miconazole are not efficacious either. Posaconazole, a newer extended spectrum oral azole has demonstrated in vitro and in vivo activity against zygomycetes, and has been used as salvage therapy for 24 patients with zygomycetes infections who were intolerant of, or whose infections were resistant to, standard antifungal therapy.[72] Surgical debridement should be considered, based on area of involvement, and sequential debridements may be necessary to control spread. Frozen-section guided debridement has been advocated to assure adequate margins.[73] Reconstructive surgery may also be necessary once the infection has been cleared. Intense management of the underlying predisposing cause of infection is also a marker for successful therapy. In diabetics with reversible acidosis, recovery rates are higher than in those patients with underlying malignancy and immunosuppression. Immunosuppressive drugs should be reduced in dosage or discontinued if possible during attempts to control the infection.

Hyperbaric oxygen therapy should be considered as adjunctive therapy, and does not replace adequate antifungal therapy. There are no clinical data that might suggest a specific treatment pressure to use in the setting of Zygomycosis infection. It would be appropriate to commence

hyperbaric oxygen treatments early in the course, rather than as salvage therapy, in the 2.4 to 3.0 atm abs range of pressures, twice a day during the acute phase of the illness, to enhance the immune response to the fungal hyphae and protect borderline ischemic areas from progression of ischemia to necrosis. Many of the successful cases have been treated with up to 30 treatments, although there are no controlled studies that would suggest a specific treatment course or pressure. Due to the rarity of the infection, it is unlikely that a prospective controlled trial could ever be done at a single institution, and thus adequate data would likely require multi-center studies, controlling for intent-to-treat vs. salvage therapy timing of hyperbaric therapy, depth and duration of treatments, as well as extent of infection at time of diagnosis, number of debridements necessary, and category of predisposing factor, along with other standard parameters.

Evidence-Based Review

From a physiological viewpoint, all necrotizing soft tissue infections should benefit from hyperbaric oxygen treatments, considering all the physiologic steps that enhance host response to infection.

Although some authors claim that hyperbaric oxygen therapy remains controversial for use to treat necrotizing fasciitis, due to lack of prospective, randomized, blinded or double blinded controlled studies comparing hyperbaric oxygen treatments vs. no hyperbaric treatments, other authors have concluded that the improvements in morbidity and mortality compared to historical morbidity and mortality data, would make it unethical to perform such randomized clinical trials on patients, as it would deny a well-substantiated adjunctive treatment for a disease with a high rate of morbidity and mortality with generally few risks of complications from the treatments. There are in fact no prospective randomized, controlled trials either, using just surgery, or surgery and antibiotics, for necrotizing fasciitis, either, yet these interventions are widely used without question, based on retrospective studies and clinical series' as well. Thus the argument against using hyperbaric oxygen therapy because of a lack of randomized controlled trials[74] can not be seriously entertained. There are numerous clinical series that do show reduced morbidity and mortality when hyperbaric oxygen has been used for numerous forms of necrotizing fasciitis, including Fournier's Gangrene. This would make it a highly recommended adjunct to antibiotics and surgical debridements.

For necrotizing fasciitis related to the Zygomycoses, the very high incidence of morbidity, disfigurement and mortality in a population that is overall usually quite compromised, leads to the conclusion that early institution of hyperbaric oxygen therapy is indicated, when the diagnosis is made, rather than waiting until signs of failure to respond to what is seemingly standard therapy, and then attempting to use hyperbaric oxygen treatments in a salvage mode, when the chances of success would seem less likely. Numerous case reports of success, even when hyperbaric oxygen treatments have been used in a salvage mode, support this recommendation. The disease process, which involves angio-invasive infection leading to ischemia, infarction, and extension of infection, appears to be one that hyperbaric oxygen treatments interfere with.

Utilization Review

Twice-a-day hyperbaric oxygen treatments during the acute phase of necrotizing soft tissue infections are advised, until extension of necrosis has been halted. Due to the natural history of often relentless progression, and undetected foci of necrosis, these treatments would then be followed by once daily treatments, over an extended period, until the infection is well-controlled, which may take up to 30 treatments. Utilization review should be requested after 30 treatments.

Cost Impact

Under a DRG system, where reimbursement is made on a daily basis, no matter what treatments are given, the cost of the treatments would be borne by the hospital while the patient is an inpatient. Due to the life-, limb-, and tissue-threatening aspects of these infections, the cost impact of hyperbaric treatments is justified. Adjunctive use of hyperbaric oxygen therapy may reduce the length of hospital stay and the number of procedures needed to attain infection control.

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