Prevention of Aminoglycoside-induced Hearing Loss

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Abstract. This review discusses the current problem of ototoxicity associated with the worldwide use of aminoglycoside antibiotics. Pathology and pathophysiology of cochlear and vestibular damage have long been recognized as a preferential destruction of hair cells beginning in the cochlea with the outer hair cells of the lower turns. This is accompanied by high frequency hearing loss progressing to lower frequencies during and even after treatment with these drugs. A novel hypothesis of the underlying biochemical mechanism is based on the formation of free radicals by an aminoglycoside-iron complex. A protective treatment against aminoglycoside-induced hearing loss by co-administration of iron chelators has been successfully documented in guinea pig. This pharmacological intervention does not change serum levels of aminoglycosides nor their antibacterial efficacy. Since iron chelators are established therapeutic drugs, the proposed treatment should lend itself to clinical application. Aminoglycosides, long established for their high efficacy and low cost, may thus continue to play an important role in combating infectious diseases. (Keio J Med 46 (3): 115-119, September 1997)

Key words: ototoxicity, gentamicin, free radicals, protection, iron chelators

Introduction

Drugs used in treatment of infections and diseases frequently have unwanted side effects. Such side effects may range from minor transient discomfort to the patient to potentially severe damage to specific organs and their function. In the latter case, it is imperative to weigh the benefits of a drug against its potential toxicity and to consider alternative treatment. There are, however, several classes of drugs with known severe side effects that remain in clinical use because of their compelling benefits. The aminoglycoside antibiotics fall into this category. The first compound in this class, streptomycin, was discovered by Waksman in 1944 and was the first successful therapy against tuberculosis. Its toxicity to the kidney and the inner ear was described one year later but streptomycin itself, and the subsequently introduced aminoglycosides (including neomycin, gentamicin, kanamycin, and sisomicin), have remained in clinical use to this day. In fact, aminoglycosides are such efficacious and inexpensive broad spectrum antibiotics that they are the most commonly used antibiotics worldwide.

Toxicity of Aminoglycosides

All aminoglycoside antibiotics display adverse side effects to the kidney and the inner ear. The relative toxicity to the kidney, the vestibular system or the cochlea varies with the specific aminoglycoside. In general, the nephrotoxicity is reversible while the ototoxicity results in permanent damage to hearing, or balance, or both. Tinnitus or decreased sensitivity of sound detection are frequent early symptoms of cochlear damage. Vestibular toxicity can be associated with vertigo, nausea, ataxia or nystagmus. The pathophysiology of ototoxicity has been well documented in several species of experimental animals and these observations agree well with findings from human temporal bones. Loss of hearing or balance is caused by degeneration of the hair cells of the cochlea and the vestibule, respectively. In the organ of Corti, hair cell loss begins with the outer hair cells in the basal turns and progresses towards the apex. The concomitant audiological symptom is a high frequency hearing loss which may later extend to lower frequencies.
Incidence of Toxicity

The incidence of ototoxicity varies with the type of aminoglycoside and is generally below 20%. Controlled clinical studies of gentamicin have determined an incidences of 6 to 16% hearing loss and 9 to 15% impairment of vestibular function. However, one study even reported an 82% incidence of hearing loss (55 of 67 patients) in tuberculosis patients receiving kanamycin. In addition, genetic susceptibility, nutritional status, noise exposure, gender, age, or pre-existing auditory or vestibular dysfunction have been discussed as risk factors in the ototoxicity of aminoglycosides.

The current problem of aminoglycoside ototoxicity is aggravated in many developing countries where the use of these drugs is widespread primarily for economic reasons and also largely unrestricted. In southeastern China, for example, two-thirds of the cases of deaf-mutism were ascribed to the use of aminoglycosides.

Despite these well known side effects, an increased use of aminoglycoside antibiotics can be expected. Opportunistic infections associated with acquired immunodeficiency syndrome (AIDS) have now resulted in a worldwide resurgence of tuberculosis, which is spreading to large segments of the general population. The World Health Organization projects tuberculosis to become the most devastating killer disease of the decade. The occurrence of resistant bacteria necessitates multi-drug regimens, which may include aminoglycosides. Given this scenario it seems more urgent than ever to prevent the toxic side effects of aminoglycoside antibiotics.

Prevention of Ototoxicity

Attempts to protect against the ototoxic side effects of aminoglycosides through pharmacological intervention began as soon as these side effects were known. However, such studies remained inconclusive and no treatment has been accepted in the clinic to date (for a summary of early studies, see; for a recent discussion, see). Most attempts at protection employed sulfhydryl compounds, radical scavengers and antioxidants, including traditional medicines and natural products but studies frequently reported contradictory findings. For example, while the free radical scavenger WR2721 was found to be effective against kanamycin ototoxicity, the scavenger N-acetyl cysteine was ineffective. Likewise, glutathione, a natural antioxidant, protected in one study but not in another. Such contradictory results may be due to complex mechanisms of toxicity and uncontrolled variables in the experimental design. At least one such confounding variable was elucidated by our laboratory when we showed that the efficacy of glutathione to attenuate gentamicin-induced hearing loss depended on the nutritional state of the animal. When animals were sick or undernourished, their natural defenses were compromised and ototoxicity was aggravated. In this case, glutathione was an effective protectant. In contrast, such dietary supplementation did not change the course of ototoxicity in healthy animals.

A further caution in the interpretation of therapeutic intervention is the potential toxicity of the prospective treatment and the interaction of the protectant with serum levels and antibacterial efficacy of the drugs. Examples of these complications can readily be found in previous attempts to ameliorate aminoglycoside toxicity. The compound 2,3-mercaptopropanol, once suggested as an antidote, was later determined to be ototoxic itself. In another case, hyperglycemic rats showed dramatic resistance to kanamycin ototoxicity but were found to excrete the drug considerably more rapidly than normal animals. Finally, poly-L-aspartate had been extensively studied as a protectant against nephrotoxicity and was also found to inhibit the cochlear toxicity of gentamicin in guinea pigs. This compound, however, also eliminated the antibacterial activity of gentamicin.

Mechanism of Ototoxicity

While such confounding factors may have contributed to inconclusive results in the past, a rational therapy was also hampered by the lack of a viable hypothesis of aminoglycoside ototoxicity. This recently changed when our laboratory was able to delineate a hypothesis on the basis of extensive in vitro and in vivo studies. The key to this hypothesis is the demonstration that aminoglycosides act as iron chelators and promote the formation of reactive oxygen species (free radicals). The development of this theory was prompted by the initially surprising finding that aminoglycosides by themselves are not toxic to isolated hair cells but require prior activation to become cytotoxic. An “activated” gentamicin, capable of catalyzing oxidation/reduction reactions was then found in the form of a gentamicin-iron chelate. Metal binding is a known mechanism in both drug effectiveness and toxicity. If iron is the chelated metal, the resulting complex will promote free-radical reactions. For example, the antineoplastic glycopeptide bleomycin requires the presence of iron for its therapeutic activity as a deoxyribonucleic acid (DNA) cleaving agent. However, free radicals can oxidize a variety of cellular targets including lipids, proteins and DNA by virtue of an unpaired electron leading to tissue damage. Reactive oxygen species, including superoxide and hydroxyl radicals, are well-documented mediators of tissue injury and have been implicated in a number of adverse drug
Free radicals are produced in all cells as a normal byproduct of metabolism, and are inactivated by a variety of enzymatic and non-enzymatic radical scavengers including superoxide dismutase, catalase, glutathione peroxidase, glutathione, vitamin C and vitamin E. If the balance between free radical production and scavenging is disturbed by external factors such as a redox-active drug, tissue damage may occur. The gentamicin-iron chelate is such a redox-active species and will produce free radicals both in vitro and in intact cells. Furthermore, the strong binding of aminoglycosides to phosphoinositides may enhance this radical formation by bringing the drugs in close proximity to arachidonic acid, which is a preferred substrate for free-radical production by gentamicin.

**Therapeutic Prevention**

Based on this hypothesis we can propose an effective therapeutic intervention against aminoglycoside ototoxicity. Since different radical species may be formed, no single radical scavenger may be completely effective. Therefore, preventing the formation of the initial aminoglycoside-iron complex may be the most promising intervention. This can be accomplished by the administration of other iron chelators which compete with aminoglycosides for available iron and thereby reduce the concentration of the aminoglycoside-iron complex. Indeed, co-administration of deferoxamine (DFO) and dihydroxybenzoic acid (DHB), both well known iron chelators, drastically reduced gentamicin ototoxicity in guinea pig. When mannitol, a hydroxyl radical scavenger and weak iron chelator, was additionally administered, the gentamicin-induced threshold shift was essentially eliminated. The effectiveness of this treatment is not limited to gentamicin but extends to other aminoglycosides and to both their vestibular and cochlear damage. Essential for prospective clinical application, the serum level of gentamicin and the antibacterial activity of gentamicin were not influenced by the co-administered drugs.

It is interesting in this context to consider a recent suggestion that N-methyl-D-aspartate (NMDA) antagonists limit aminoglycoside-induced hearing loss. Such a suggestion apparently fits well with documented polyamine-aminoglycoside antagonism in many systems, including competition for uptake in the inner ear and inhibition of cochlear ornithine decarboxylase. However, in the study of NMDA antagonists, the two protective compounds dizocilpine and ifenprodil were given as their maleate and tartrate salts, respectively. Both maleate and tartrate have metal chelating properties and, therefore, the suggested protection may be due to metal chelation rather than polyamine antagonism. This question needs to be resolved with appropriate control studies.

**Clinical Application**

The proposed preventive therapy against aminoglycoside ototoxicity should be clinically applicable, because iron chelators have been well established in clinical studies and found to be safe. DFO is currently the drug of choice in the treatment of chronic iron overload, for example in ß-thalassemia. It should be cautioned, however, that DFO has some intrinsic neurotoxicity to both the retina and inner ear although this appears mostly in association with very high doses. Nevertheless, alternative iron chelators may be preferable. The other drug so far tested as a protectant by our laboratory, DHB, has undergone preliminary clinical trials and was found to be essentially non-toxic. Furthermore, the combined treatment with gentamicin and DHB also increased survival rates in septic rats as compared with gentamicin alone suggesting additional potential benefits of co-therapy. Finally, both chelators may also attenuate aminoglycoside-induced nephrotoxicity.

While efficacy and safety should always be a primary concern in medical practice, the cost factor will play a decisive role in developing countries. Acceptance of a "safe" aminoglycoside treatment protocol requires that the treatment is simple and does not substantially increase the cost of treatment. Future studies of prospective therapeutics need to take this economic reality into consideration.

In summary, in vitro experiments and animal studies have established a new hypothesis of aminoglycoside ototoxicity. It is based on the ability of aminoglycosides to chelate iron and promote the formation of free radical reactions. From this basis, a protective treatment with competing iron chelators has been designed and successfully documented in the guinea pig. Aminoglycosides, long established for their high efficacy and low cost, may thus continue to play an important role in combating infectious diseases.

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