Vancomycin Enhances the Ototoxic Properties of the Aminoglycoside Antibiotic Gentamicin

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Background

- Sensori-neural hearing loss and tinnitus are well documented side effect of aminoglycosides
- Selective cochlear hair cell destruction
- Certain aminoglycosides can be vestibulotoxict
- Theorised due to the generation of ROS
- Higher concentrations → to cell death
- Lower Concentrations → membrane blebbing and phospholipid externalisation
- Both processes associated with apoptosis
Figure 2. Taken from Experimental Project Data, illustrating the difference between hair cells which have not been treated with neomycin (left panel) and those that have (right panel)
Vancomycin Ototoxicity

- Vancomycin is also listed in the BNF as causing sensori-neural hearing loss and tinnitus.
- Evidence is less substantial, most being case based.
- 3 *in vivo* studies done previously, generating contentious results.
- No *in vitro* evidence for ototoxicity.
Aims of Project

- To assess the ototoxic properties of vancomycin in comparison to aminoglycoside induced toxicity.
- Also to examine if vancomycin enhances the ototoxic properties of gentamycin.
- In vivo studies have contentious, and thus this project will examine the direct in vitro effects of the compounds on cochlear hair cells.
**Methods - Preparation**

- Cochlear Hair cell cultures were produced from 2 day old mouse pups under the appropriate techniques outlined in Richardson and Russel in 1987.

- Cultures treated with varying concentrations either vancomycin, aminoglycoside or control solution (saline solution) for 72hrs.
Methods-Techniques

- **Phalloidin Fluorescence:**
  - Naturally occurring poison in death cap mushrooms bound to a fluorophore
  - Stains F-Actin in stereociliary bundles was used to assess hair bundle morphology & living vs dead hair cells

- All cultures were analysed qualitatively:
  - Living/Functional cells demonstrate the characteristic V-Shaped stereocilia under fluorescence
  - Damaged tissue demonstrate flattening of the stereocilia or poor fluorescence
  - Dead tissue doesn’t fluoresce
Vancomycin (2 mMol and 5 mMol)
Discussion - Vancomycin

• A total of 4 phalloidin fluorescence experimental series were performed, with 28 different cochlear cultures examined

• Cultures treated with 0.5mMol, 1mMol and 2mMol show no significant differences to cultures treated with control solution, with normal morphology and no significant cell death

• Only 5mMol cultures showed abnormal hair bundle morphology, but this could be due to alterations in pH medium

• However, this is an extremely high vancomycin level and could not be replicated physiologically

• Treatment with both aminoglycoside and vancomycin did show significant increases in hair cell loss from cultures treated with a similar concentration of aminoglycoside alone.
Conclusions

• *In Vitro* evidence that Vancomycin does not itself cause damage to hair cells

• Limitations: mouse model, external random error, also an assessment of structure rather than functionality

• Further work: Observational patient study of vancomycin ototoxicity to review concurrent aminoglycoside prescription, Scanning Electron Microscopy data for Vancomycin

• Also data needs validation via further repetition

• However, this project generates evidence that vancomycin neither causes hair cell death or apical surface damage

• There is evidence that vancomycin does enhance aminoglycoside ototoxicity
Many Thanks for Listening

Any Questions?
References

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- Russell IJ, Richardson GP. The morphology and physiology of hair cells in organotypic cultures of the mouse cochlea. Hear Res. 1987 Nov;31(1):9–24