A holistic approach to the safety assessment of exploratory drug targets

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INTRODUCTION

Safety liabilities of a drug and - more importantly - its associated target are often not placed in a broad enough context, which contributes significantly to the high drug attrition rate in the pharmaceutical industry. There is thus an evident need for an efficient and holistic target safety evaluation workflow that incorporates all aspects of target safety. At TNO – in collaboration with a pharmaceutical company – we have uniquely combined all necessary disciplines to develop a holistic workflow that collects, integrates and interprets all relevant information necessary to assess and rank potential toxicity issues of an exploratory target. The resulting workflow – benchmarked with HSP90 inhibition as demonstrator – serves as a blueprint for any company embarking on target nomination and validation.

FROM CONCEPT to WORKFLOW

HSP90 Target Safety Assessment

1a. HSP90 characterization

Biosysntetic Function

HSP90 is a unique molecular chaperone that functions as a homodimer. The complex pedesols within the proper folding of specific target proteins by providing structural stability, which is essential for the proper functioning of the cell. HSP90 is involved in numerous pathways key roles in the protein-signaling pathway, protein folding, protein degradation, and cytoskeletal interactions and antigen presentation.

Desired therapeutic effect

Promoting/reducing CNS-diseases

Toxins

- HSP90 alpha
- HSP90 beta

Homologues (‘isofoms’)

- GRP94
- TRAP1

Expression profile

Ubiquitin

Ligand binding site

- ATP (binding site, N-terminal high affinity, R•-xy, arg-gly-phenylalanine)
- ATP (binding site, C-terminal low affinity, R•-xy, his-lys-argin

Mechanism of Action

Inhibition of HSP90

Downstream effect

Up-regulation of HSP70

1b. Most advanced structural compound classes

Cluster 1: (GSK-3b)
Cluster 2: (DAPI)
Cluster 3: (TRAP1)
Cluster 4: (grp94)
Cluster 5: (HSP90)

Therapeutic indication of HSP90

1c. Isoform selectivity

Overlay of HSP90 (LYT, blue), GRP94 (LYT2, orange), TRAP1 (4KID, green), displaying high conservation of the N-terminal domain and inhibitor binding site (left). However, the ligand-induced fit of HSP90 residues 104-311 (red) allows for isoform-selective inhibition (right).

2. Systems biology

HSP90 Pathways - Pathologies

Mechanistic toxicity:

HSP90 related-cadidocytosis is partly due to the role of the alpha isoform in the proper folding and trafficking of NM23.

Right: Strikingly potent showing protein-protein binding

3a. Major toxicities HSP90

Cluster

- Cardioxicity
- Hepatotoxicity
- Neurotoxicity
- Immunotoxicity
- Renal toxicity
- Gastro-intestinal toxicity
- Hematotoxicity
- Ocular toxicity
- Osteoclast formation
- Pulmonary & skeletal muscle effects
- Reproductive effects

3b. Role of related proteins

- GRP94 function
  - Cell defense mechanisms
  - Apoptosis
  - Inflammatory response
- TRAP1 function
  - Mitochondrial integrity & calcium homeostasis
  - Endoplasmic reticulum stress
- HSP70 function
  - Proteasomal activity

4. Toxicity profile

Risk ranking of HSP90-related toxicity based on severity, frequency, evidence and Alzheimer's patient co-morbidities:

Cardiotoxicity & Hepatotoxicity

Retinal, gastro-intestinal & renal toxicity

Immune toxicity, CNS effects

Osteoclast formation

Pulmonary & skeletal muscle effects

Reproductive effects

5. Risk mitigation

Risk mitigation encompasses:

- Specific screening assays for the identified (mechanistic) toxicities
- PK analysis (retinal toxicity)
- Assessment of reactive metabolite formation (liver toxicity)
- Optimization of compounds towards isoform-selective inhibitors

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