

## ALS: ARE PRECLINICAL ANIMAL STUDIES PREDICTIVE AND NECESSARY FOR DRUG DEVELOPMENT? - PRO

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Disease modifying treatments for amyotrophic lateral sclerosis (ALS) are urgently needed since presently only one drug, riluzole, interferes positively with the natural history of the disease and its effect remains limited. Since 1994, a transgenic animal model exists for the disease which is based on an established etiological factor – mutations in the SOD1 gene. During the past decade, the field of translational research in ALS/MND was largely built on the use of this transgenic animal model (Gurney et al., 1994, Rothstein 2003). Caused by a multifold overexpression of the gene, the transgenic animal develops motor deficits and dies from respiratory failure - both caused by loss of anterior horn cells. Initially, there was much hope in the field that this model could greatly facilitate efficient drug development in ALS. This hope was seemingly fulfilled by the positive effects of riluzole, in both experimental mice and men. Until now, more than 100 (published and unpublished) compounds were tested with apparent success in this animal model; however, many attempts to translate these experimental studies into therapy in humans failed. Therefore, skeptics doubt whether animal models are of any use for drug development in ALS. However, in my view this black and white thinking does not see the reality of preclinical drug development. To reduce the high costs of clinical drug development, rather than abandoning them, improvement of animal studies is necessary (not only in translational research in ALS). How can we achieve this goal ?

### 1. Improvement of the methodology of transgenic animal studies

To improve the success rate of preclinical research and to decrease unjustified investments, clinical and preclinical researchers met in Holland in March 2006 in a workshop in order to define guidelines for the use of standard methods for drug testing in mouse models. The workshop members agreed on the following three principles on which the more detailed guidelines were built (Ludolph et al., 2007):

- I. It was felt that it was necessary to develop guidelines for preclinical research since a number of drug-finding studies are found by many in the field to be unreliable for translational research, the step from mice to men. The guidelines are to be developed to improve the quality of the studies and consequently save resources for both, preclinical and clinical studies.
- II. It was decided to consider "proof of concept" and "preclinical studies" separately. A *proof of concept* study has the goal to elucidate the mechanism of the disease, may it be biochemical or physiological. Such a study may use a drug as an investigational tool. A *preclinical study* has the primary goal to develop a drug for use in humans and must underlie more rigid methodological considerations..
- III. It was expressed that the presence of one model representing only a single etiological factor of ALS/MND is not satisfying. The development of more models based on other etiological factors than SOD mutations is warranted, in particular to have the opportunity to validate more accurately the targets of disease-modifying drugs.  
In the future, these thoughts and their consequences are likely to be employed in a similar fashion to translational research in other neurodegenerative diseases.

### 2. To distinguish etiological factors and pathogenetic processes common to the disease of the motor neurons

Historically, the definition of many neurodegenerative diseases was built on the analysis of clinical and neuropathological phenotype and their relation (Charcot's method). If we are to develop therapies for all patients with motor neuron diseases, specific etiological factors (f. e. SOD mutations, dynactin mutations) must be distinguished from common factors of the pathogenesis of all models. Therefore models must be compared and pathogenetic factors ("targets") must be validated. This approach reflects the concept of selective vulnerability ("Pathoklise") as presented by the Vogts in the early 20<sup>th</sup> century. However, the Vogts views should be modified by the concept of neuroplasticity which implies the presence of major adaptive changes in the damaged nervous system. This process could be dependent from a number of factors, including age of the organism and potency of the etiological factor.

### 3. Improved understanding of the pathogenesis and development of biomarkers in mice and men

Not astonishingly, recent experimental and clinical results suggest that prevention may be different from treatment strategies of ALS. Experimentally, the results of Don Cleveland's group (Boillee et al., 2006) currently have a major influence on thought development in the field of ALS/MND. In brief, his group showed that cell death in models of ALS/MND is non cell-autonomous. More specifically, experimentally the onset of the disease is determined by toxic influences inherent to the motor neurons themselves whereas progression of the clinical disease may be more dependent from neuroinflammation. Seemingly complementary to these results are studies of the therapeutic and preventive capacity of vitamin E. Whereas intervention during the clinical course of the disease had no effect in two independent studies, retrospective analysis of two cohorts (Ascherio 2004, Veldink 2005) showed that increased intake of moderate dosages of vitamin E decreased the risk of ALS/MND by the amount of 63 %. Based on these insights, biomarkers are to be developed in CSF and blood of experimental animals and men which reflect the integrity of the neuroaxonal system (f. e. Tau), astrocytes (f.e. S100 $\beta$ ), microglia (f.e. cytokines) and other components of the system (for example endothelial cells).

In conclusion, in my view frustration with the results of pharmacological translational research in ALS is understandable, but should not lead to overreactions. Experimental preclinical research is urgently needed and its improvement is necessary. The goals can be achieved by

- a. improvements of the methodology of preclinical research, in particular in animals
- b. comparing the pathogenesis of the animal models for motor neuron diseases (target validation) and
- c. the development of cell-specific biomarkers.

#### References:

- Ascherio A, et al. Vitamin E intake and risk of amyotrophic lateral sclerosis. *Annals of Neurology* 2005; 57:104-10.  
Boillee S, et al. Onset and progression in inherited ALS determined by motor neurons and microglia. *Science* 2006;312:1389-92.  
Gurney ME, et al. Motor neuron degeneration in mice that express a human Cu/Zn superoxide dismutase mutation. *Science* 1994;264:1772-1775.  
Ludolph AC, Bendotti C, et al. Guidelines for the preclinical evaluation of pharmacological active drugs for ALS/MND: Report on the 142<sup>nd</sup> ENMC international workshop. *Amyotrophic Lateral Sclerosis* 2007;8(4):217-23.  
Rothstein JD, et al. Of mice and men: Reconciling preclinical ALS mouse studies and human clinical trials. *Annals of Neurology* 2003;53:423-426.  
Veldink JH, et al. Intake of Polyunsaturated fatty acids and vitamin E reduce the risk of developing ALS. *J Neurol Neurosurg Psychiatry*. 2007 78(4):367-71. Erratum in: *J Neurol Neurosurg Psych* 2007;78(7):779.