OF MICE AND MONKEYS:

Choosing the Ideal Model for Rheumatoid Arthritis

Rheumatoid arthritis (RA) is an autoimmune disorder affecting up to 1% of the population, associated with significant morbidity and mortality if not treated aggressively. Justifiably, it has been a target of great interest to biotech and pharma companies because, although there are now many drugs available, no one single drug or class of drug is universally effective and all have toxicities that can limit their use.

A major limitation in the genesis of new therapeutic strategies for many human diseases, including RA, is the absence of practical and accurate animal models. Some toxicities can be identified in small animals, e.g., mice, rats, but these do not necessarily predict all human issues. Therapeutic interventions into mouse and rat models may not predict efficacy in the human disease: the intended target for human RA may differ from the animal analog, which may not be affected by the therapeutic molecule intended for RA. Non-human primates (NHP) would be the logical next step in developing a model of RA. NHPs may represent a better choice than rodents: NHPs are easier to manipulate and have an immune system much more like that of humans.

The collagen-induced arthritis (CIA) model has been established in rhesus macaques, marmosets and cynomolgus monkeys; these models mimic human RA in many ways and are thought to be more indicative of human disease than rodent CIA models, e.g. mouse, rat. However, working with rhesus monkeys has limitations: their large size means larger amounts of test substance is required and animal housing costs are higher; a major limitation is that the incidence of CIA in rhesus monkeys is only about 60%. Marmosets are evolutionarily farther away from humans than are rhesus monkeys and sufficient numbers of animals may not be available for routine adoption of this model into routine preclinical drug development.

PharmaLegacy has developed a practical CIA model of human RA using cynomolgus monkeys, based on outbred strains. A high incidence of CIA is achieved (>80%) by using a two-step immunization strategy. The average cynomolgus monkey is much smaller than a rhesus monkey, offering a cost saving advantage in housing; taken together the smaller amount of test substance used per animal and the higher incidence of CIA mean less test substance is required. Finally, there are sufficient numbers of these NHPs to make the model practical. The evolutionary distance from cynomolgus monkeys to humans is much less than that of marmosets, making the cynomolgus monkey model a more likely mimic of human RA and its pathogenesis. Supporting the likelihood that this new model will be relevant to the study of human RA are the following facts: The Multi-Biomarker Disease Activity test used to determine activity of human RA also correlated with disease progression in the cynomolgus monkeys; the mRNA profile in affected cynomolgus monkey joints revealed elevated levels of IL-1β, IL-6, GM-CSF and RANKL, as well as IL-17A; and anti-IL-6 treatment inhibited IL-1β/IL-6 inflammatory pathways, as well as suppressing the severity of arthritis, just as IL-6 blockade does in human RA.

Thus, the cynomolgus model merges the advantages of rhesus macaques (close genetic relationship to humans, availability) and of marmosets (high disease onset rate, less test substance needed and reduced housing costs), without the limitations intrinsic to either of these models.

PharmaLegacy has Non-Human Primate Models covering many disease areas, including:

- Rheumatoid Arthritis
- Lung Inflammation
- Inflammatory Bowel Disease (IBD)
- Delayed Type Hypersensitivity (DTH)
- Experimental Autoimmune Encephalomyelitis (EAE)
- Thrombosis
- LPS-Induced Systemic Infection
- Chemotherapy Induced Mucositis
- Many more + bespoke models available upon request

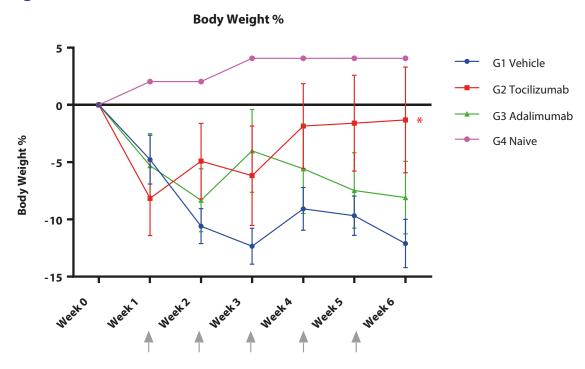## Evaluating the efficacy of tocilizumab and adalimumab in collagen induced arthritis in cynomolgus monkeys

Study design:

Group	Test article [all biologics]	N *	Route + Schedule
1	Vehicle (saline)	6	Slow IV infusion, QW for 5 wk
2	Tocilizumab #	6	Slow IV infusion, QW for 5 wk
3	Adalimumab #	6	Slow IV infusion, QW for 5 wk
4	Naïve, no CIA	1	-

* Number of animals actually treated in the study
\# Tocilizumab (Actemra®) and Adalimumab (Humira®)
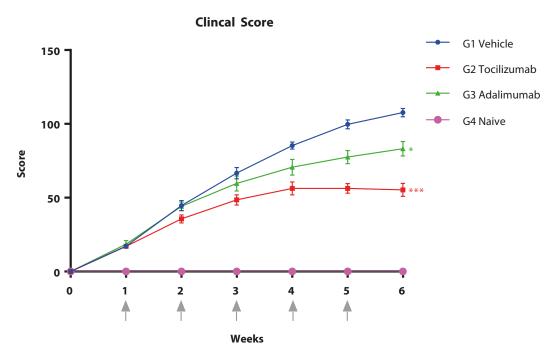
Results:

• Body weight:

Body Weight %

* p<0.05, G2 & G3 *vs* G1 (Repeat measurement ANOVA/Bonferroni's)
Note: Arrows were the delivered doses.

Tocilizumab treatment significantly prevented the loss of body weight in cynomolgus monkeys and adalimumab showed a trend to prevent body weight loss compared with vehicle group.
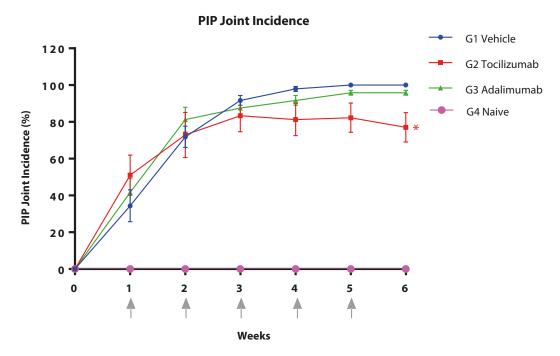
• Clinical arthritis score:

Clincal Score

* p<0.05 and ***p<0.001, G2&G3 *vs* G1 (Repeat measurement ANOVA/Bonferroni's)
Note: Arrows were the delivered doses.

The clinical arthritis score in both tocilizumab and adalimumab treatment groups decreased significantly in comparison with vehicle group.
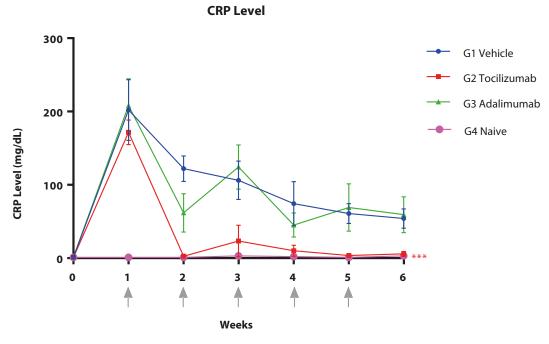
• PIP joint incidence:

PIP Joint Incidence

* p<0.05, G2&G3 *vs* G1 (Repeat measurement ANOVA/Bonferroni's)
Note: Arrows were the delivered doses.

Tocilizumab treatment significantly decreased PIP joint incidence in comparison with vehicle group.
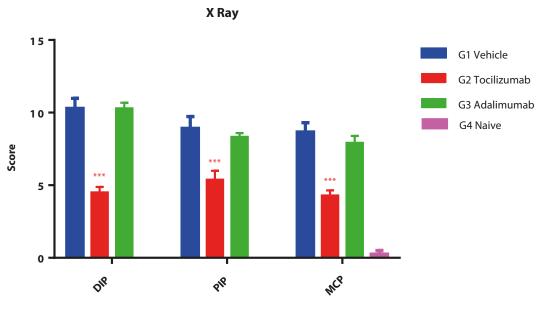
• CRP level:

CRP Level

*** p<0.001, G2&G3 *vs* G1 (Repeat measurement ANOVA/Bonferroni's)
Note: Arrows were the delivered doses.

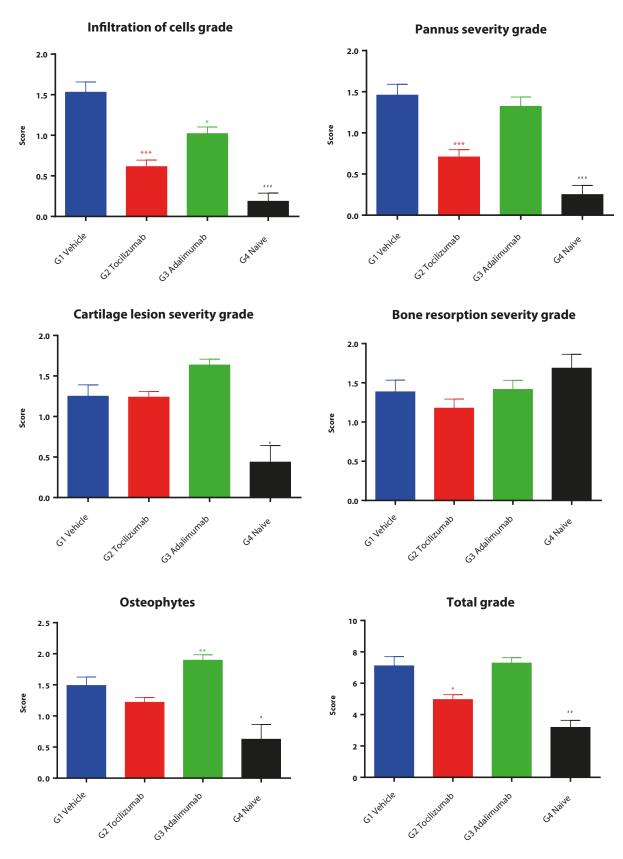Tocilizumab treatment significantly decreased serum level of CRP.

• X-ray examination:

X Ray

*** p<0.001 G2&G3 compared with G1 (Krusk al-Wallis/Dunn's)

Tocilizumab treatment significantly improved joint distortion in all DIP, PIP and MCP.

• PIP histopathology:

Infiltration of cells grade

Pannus severity grade

Cartilage lesion severity grade

Bone resorption severity grade

Osteophytes

Total grade

Tocilizumab treatment significantly decreased inflammatory cell infiltration and improved the PIP histopathology.

Being Correct is Everything

To advance a compound from discovery to clinical, or to halt its development, is a huge and costly decision. For the benefit of you and your company, and for the well-being of patients in need of treatment, that decision needs to be based on correct information.

PharmaLegacy has more capability to provide rich, correct answers to pharmacological questions due to our huge repository of in vivo models, the rich experience of our company and scientists, our intense focus on pharmacology, and our proprietary technological platforms.

There's a lot on the line. Let PharmaLegacy get the correct answers.

Quick Facts:

- Over 300 validated animal models of disease spanning over 40 different diseases
- Scientific staff average over 15 years of pharmacology experience
- FDA Part 11 compliant
- 45,000 ft2 facility with 22,000 ft2 of SPF and conventional vivarium to house 10,000 rodents and large animals
- On-site capacity for up to 350 non-human primates
- Partnerships with three premium non-human primate centers having capacity for 30,000 monkeys (Cynomolgus and Rhesus)
- Capacity to run 200 animal studies concurrently while strictly following AAALAC and ILAC guidelines
- Research data is electronically managed by BioBook (IDBS, UK)
- Web-based live video streaming allows remote monitoring of operations from any location worldwide
- Operations structured for maximum protection of clients' work and intellectual property
- 24/7 access to PharmaLegacy representatives

www.pharmalegacy.com

Expect More from Pharmacology

No animal study can predict with certainty how a therapy will perform in human subjects, yet that is what you need them to do. You need to get as much information from them as possible. Due to study design, the models and technologies available, or lack of proper expertise, most preclinical, in-vivo pharmacological studies are not providing as much information as they could be. These missing insights can derail a pipeline. Missed opportunities are not what you should expect from pharmacology CROs. You should expect more.

Now you can

You can expect more from PharmaLegacy because PharmaLegacy has more: more models, more experience, and more focus. We have over 300 validated animal models, including non-human primate models and many humanized rodent models, spanning over 40 different diseases in the fields of cancer, auto-immunity, inflammation and bone. Our staff has an average of 15 – 20 years of pharmacology experience, and most of them have been with PharmaLegacy for more than 5 years.

When you search for a pharmacology CRO, set your expectations high.
Contact PharmaLegacy and let's discuss how we can exceed them.