

## Immunological Evaluation of BIOCELL® INTRASHIEL® Shell in Female B6C3F1 Mice

### VII. Conclusions

These studies were conducted for McGhan Medical Corporation by [REDACTED] to determine the potential of the INTRASHIEL® Shell Test Article to compromise the immune system. In female B6C3F1 mice implanted with INTRASHIEL® Shell Test Article for 28 days, no biologically significant effects were observed on standard toxicological parameters including body weight, body weight gain, spleen weight, thymus weights, gross pathology, thymus histopathology, or hematological parameters.

Overall, no significant differences were observed between the sham control animals and the animals implanted with the INTRASHIEL® Shell Test Article in the immunological studies. Exposure to the INTRASHIEL® Shell Test Article did not result in significant changes in the antibody-forming cell (AFC) response to the T-dependent antigen, sheep erythrocytes, when evaluated as specific activity (AFC/10<sup>5</sup> spleen cells). While a significant increase in total spleen activity (AFC/spleen) was observed in the high dose animals implanted with the INTRASHIEL® Shell Test Article, the increase appears to be related to a low response of the sham control animals as opposed to being related to the INTRASHIEL® Shell Test Article.

There were no significant effects on the spleen B cell, T cell, or T cell subsets following exposure to the INTRASHIEL® Shell Test Article with the exception of the immature double positive (CD4<sup>+</sup>CD8<sup>+</sup>) T cells. A significant decrease (39%) was observed at the two highest dose levels of animals implanted with the INTRASHIEL® Shell Test Article. However, the double positive cells make up a very small percentage of the spleen population, usually less than 4%, and thus very slight changes in cell number can result in statistical differences, without functional relevance. Exposure to INTRASHIEL® Shell Test Article did not result in significant changes in the mixed leukocyte response (MLR) or in natural killer (NK) cell function, when evaluated either as specific activity (LU/10<sup>7</sup> spleen cells) or total spleen activity (LU/spleen).

In conclusion, the results of this comprehensive immunotoxicological evaluation demonstrate that, under the experimental conditions used, exposure to the INTRASHIEL® Shell Test Article did not adversely affect the functional ability of the immune system.

### VIII. REFERENCES

1. Bradley, S.G., A.E. Munson, J.A. McCay, R.D. Brown, D.L. Musgrove, S. Wilson, M. Stern, M.I. Luster and K.L. White, Jr. 1994. Subchronic 10 day immunotoxicity of polydimethylsiloxan (silicone) fluid, gel and elastomer and polyurethane disks in female B6C3F1 mice. *Drug and Chemical Toxicol.* 17(3), 175-220.
2. Bradley, S.G., K.L. White, Jr., J.A. McCay, R.D. Brown, D.L. Musgrove, S. Wilson, M. Stern, M.I. Luster and A.E. Munson. 1994. Immunotoxicity of 180 day exposure to polydimethylsiloxan (silicone) fluid, gel and elastomer and polyurethane disks in female B6C3F1 mice. *Drug and Chemical Toxicol.* 17(3), 221-269.

3. Luster, M.I., A.E. Munson, P. Thomas, M.P. Holsapple, J. Fenters, K.L. White, Jr., L.D. Lauer, D.R. Germolec, G.J. Rosenthal, and J.H. Dean. 1988. Development of a testing battery to assess chemical-induced immunotoxicity: National Toxicology Program guidelines for immunotoxicity evaluation in mice. *Fund. Appl. Toxicol.* 10:2-19.
4. Jorne, N.K., C. Henry, A.A. Nordin, H. Fun, M.C. Koros, and I. Lelkovits. 1974. Plaque forming cells: Methodology and theory. *Transpl. Rev.* 18:130-191.
5. Munson, A.E., J.A. McCay and W. Cao. 1991. Approaches to immunotoxicologic studies with emphasis on chemical-induced immunomodulation. *Annals of Allergy* 66:505-518.
6. Thorpe and Knight. 1984. *J. Immunol. Meth.*, 5:387.
7. Klassing, R., E. Klein, and H. Wigzell. 1975. "Natural" killer cells in the mouse. I. Cytotoxic cells with specificity for mouse Moloney leukemia cells. Specificity and distribution according to genotype. *Eur. J. Immunol.* 5:112-121.
8. Luster, M.I., C. Portier, D.G. Pait, R.J. Rosenthal, D.R. Germolec, C.E. Comment, E. Corsini, B.L. Blaylock, P. Pollock, Y. Kouchi, W. Craig, K.L. White, Jr. and A.E. Munson. 1993. Risk assessment in immunotoxicology: II. Relationship between immune and host resistance tests. *Fundam. Appl. Toxicol.* 21:71-82.
9. Bartlett, M.S. 1937. Sub-sampling for attributes. *J. Roy. Stat. Soc. Suppl.* 4:131-135.
10. Kruskal, W.H., and W.A. Wallis. 1952. Use of ranks in one-criterion variance analysis. *J. Amer. Stat. Assoc.* 47:583-621.
11. Dunnett, C.W. 1955. A multiple comparison procedure for comparing several treatments with a control. *J. Amer. Stat. Assoc.* 50:1096-1121.
12. Gross, A.J., and V.A. Clark. 1975. Gehan-Wilcoxon Test. In *Survival Distribution: Reliability Applications In Biomedical Sciences*. A.J. Gross and V.A. Clark, eds. John Wiley and Sons, New York, p.120-123.
13. Sokal, R.R. and F.J. Rohlf. 1981. *Biometry*. Freeman, San Francisco, p.226-231.6