promised development is a relief. However, the future may be more complex than commonly envisioned by positivistic reductionists.

Studies involving complex disease or disease-like test systems may not be reduced to simple integrative or 'translational' (ref. 1) research that merely seeks confirmation of *in vitro* cell and molecular data/concepts. For example, "translational" research would negate the fact that cells *in vitro* may exhibit features (physiology, pharmacology, and ultrastructure) distinct from those recorded *in vivo*[3]. Clinical and other *in vivo* studies of new bioactive molecules should include the active search for off-label compound actions, as these often lead to innovative treatments[3–5]. Thus, complex biosystems investigators remain a driving force behind the discovery of drugs and essential disease mechanisms[3–5]. This development, which is complementary to molecular medicine, is in keeping with the fallacy of the paradigm of genetic determinism for many common diseases.

A reappraisal of 'wholistic' *in vivo* research as both a validation and a discovery activity will be of tremendous importance to reductionists who now may have to adopt (explore and explain) incorrect or insufficiently researched textbook ideas of physiology and histopathology. Scientists trained in exploratory *in vivo* approaches will also speed up critical evaluations of *in vitro* research standards, to reduce the risk that they become 'segregative'. Collaborations giving equal merit to *in vitro* and *in vivo* research will vouch for the lack of delay in exploring the mechanisms involved in original *in vivo* discoveries (not forgetting iconoclastic observations). Encouraging scientists to study complex biosystems, using ever-improved physiological and histopathological methods as well as adopting the molecular techniques, is urgently needed. Indeed, the failure to understand the importance of exploratory *in vivo* approaches may profoundly slow discovery in medicine[3–5].

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1. Bell, J.I. Clinical research is dead; long live clinical research. *Nature Med.* **5**, 477–478 (1999).
2. Crooke, S.T. New drugs and changing disease paradigms. *Nature Biotechnol.* **14**, 238–241 (1996).
3. Persson, C.G.A. Centennial notions of asthma as an eosinophilic, desquamative, exudative, and steroid-sensitive disease. *Lancet* **349**, 1021–1024 (1997).
4. Wurtman, R.J. & Bettiker, R.L. The slowing of treatment discovery, 1965–1995. *Nature Med.* **1**, 1122–1125 (1995).
5. Gutterman, J.U. Clinical investigators: The driving force behind drug discovery. *Nature Biotechnol.* **15**, 598–599 (1997).

Oxidative DNA damage and embryo development

To the editor—We read with interest the findings of Parman and colleagues regarding the etiology of thalidomide-induced embryopathy[1]. The authors suggest that excess reactive oxygen species (ROS) may be involved in the teratogenic process of thalidomide. Their conclusions are based on increased levels of DNA damage in thalidomide-exposed rabbit embryos, concomitant with induction of embryonic developmental damage. Pre-treatment of the mother with the anti-oxidant α-phenyl-N-t-butylnitrone was shown to reduce embryonic dysmorphogenesis. In earlier studies, the authors presented evidence suggesting that excess ROS may be involved in the induction of phenytoin-associated congenital malformations[2], and others have suggested a role for ROS in ethanol-induced embryopathy[3].

Excess ROS may also be important in the maldevelopment seen in the offspring of diabetic mothers. This idea, has been substantiated by *in vivo* demonstrations of diminished dysmorphogenesis in the offspring of diabetic rodents given dietary antioxidative agents, such as butylated hydroxytoluene[4], vitamin E (ref. 5), vitamin C (ref. 6) and lipoic acid[7]. Pregnant diabetic mice transgenic for *Sod1* show fewer embryo malformations than non-transgenic pregnant diabetic controls[8]. It has been proposed further that oxidative stress in embryos exposed to a diabetic environment may be the result of increased levels of the isoprostane 8-epi-PGF_2 in the embryos[9]. Also, the fact that high-amplitude mitochondrial swelling in embryonic neuroectoderm of these embryos[10] is diminished by antioxidative treatment of the mother[11] suggests the presence of an embryonic ROS imbalance, with conceivable consequences for the rate of apoptosis in susceptible cell lineages in the embryo[12].

Finally, just as in thalidomide teratogenicity, the fetuses and embryos of diabetic rodents have increased rates of DNA damage[13,14], suggesting the possibility of a common teratological pathway involving altered expression of genes under the control of transcription factors sensitive to oxidative stress. Investigations of diabetic pregnancies have identified candidate genes, including catalase[15] and cyclooxygenase-2 (ref.9).

These intriguing genetic and biochemical relationships associated with teratological process(es) demand further study. The effect of excessive ROS on embryogenesis may emerge as a more general mechanism in teratogenesis than previously thought.

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1. Parman, T., Wiley, M.J. & Wells, P.G. Free radical-mediated oxidative DNA damage in the mechanism of thalidomide teratogenicity. *Nature Med.* **5**, 582–585 (1999).
2. Winn, L.M. & Wells, P.G. Phenytoin-initiated DNA oxidation in murine embryo culture, and embryoprotection by the antioxidative enzymes superoxide dismutase and catalase: evidence for reactive oxygen species mediated DNA oxidation in the molecular mechanism of phenytoin teratogenicity. *Mol. Pharmacol.* **48**, 112–120 (1995).
3. Kotch, L.E., Chen, S.-E. & Sulik, K.K. Ethanol-induced teratogenesis: free radical damage as a possible mechanism. *Teratology* **52**, 128-136 (1995).
4. Eriksson, U.J. & Simán, C.M. Pregnant diabetic rats fed the antioxidant butylated hydroxytoluene show decreased occurrence of malformations in the offspring. *Diabetes* **45**, 1497–1502 (1996).
5. Viana, M., Herrera, E. & Bonet, B. Teratogenic effects of diabetes mellitus in the rat. Prevention with vitamin E. *Diabetologia* **39**, 1041–1046 (1996).
6. Simán, C.M. & Eriksson, U.J. Vitamin C supplementation of the maternal diet reduces the rate of malformation in the offspring of diabetic rats. *Diabetologia* **40**, 1416–1424 (1997).
7. Wiznitzer, A. *et al.* Lipoic acid prevention of neural tube defects in offspring of rats with streptozocin-induced diabetes. *Am. J. Obstet. Gynecol.* **180**, 188–193 (1999).
8. Hagay, Z.J. *et al.* Prevention of diabetes-associated embryopathy by overexpression of the free radical scavenger copper zinc superoxide dismutase in transgenic mouse embryos. *Am. J. Obstet. Gynecol.* **173**, 1036–1041 (1995).
9. Wentzel, P., Welsh, N. & Eriksson, U.J. Developmental damage, increased lipid peroxidation, diminished cyclooxygenase-2 gene expression, and lowered PGE2 levels in rat embryos exposed to a diabetic environment. *Diabetes* **48**, 813–820 (1999).
10. Yang, X., Borg, L.A.H. & Eriksson, U.J. Altered mitochondrial morphology of rat embryos in diabetic pregnancy. *Anat. Rec.* **241**, 255–267 (1995).
11. Yang, X., Borg, L.A.H., Simán, C.M. & Eriksson, U.J. Maternal antioxidant treatments prevent diabetes-induced alterations of mitochondrial morphology in rat embryos. *Anat. Rec.* **251**, 303–315 (1998).
12. Forsberg, H., Eriksson, U.J. & Welsh, N. Apoptosis in embryos of diabetic rats. *Pharmacol. Toxicol.* **83**, 104–111 (1998).
13. Lee, A.T., Plump, A., DeSimone, C., Cerami, A. & Bucala, R. A role for DNA mutations in diabetes-associated teratogenesis in transgenic embryos. *Diabetes* **44**, 20–24 (1995).
14. Lee, A.T., Reis, D. & Eriksson, U.J. Hyperglycemia induced embryonic dysmorphogenesis correlates with genomic DNA mutation frequency *in vitro* and *in vivo*. *Diabetes* **48**, 371–376 (1999).
15. Cederberg, J. & Eriksson, U.J. Decreased catalase activity in malformation-prone embryos of diabetic rats. *Teratology* **56**, 350–357 (1997).