APPENDIX 1: NanoRiskCat●●●|●● Template

Literature methodology/sources of information

The following sources of information were used to fill out the NanoRiskCat●●●|●● for Carbon nanotubes:

1. Allen, B.L., Kichambare, P. D., Gou, P., Vlasova, I. I., Kapralov, A.A., Konduru, N., Kagan, V. E., Star, A., 2008. Biodegradation of single-walled carbon nanotubes through enzymatic catalysis. Nano Lett. 8, 3899-3903.
2. Kennedy AJ, Hull MS, Steevens JA, Dontsova KM, Chappell MA, Gunter JC, Weiss CA Jr. 2008. Factors influencing the partitioning and toxicity of nanotubes in the aquatic environment. Environmental Toxicology & Chemistry 27(9): 1932-1941
3. Tran CL, SM Hankin, B Ross, RJ Aitken, AD Jones, K Donaldson, V Stone, R Tantra, 2008, 'An outline scoping study to determine whether high aspect ratio nanoparticles (HARN) should raise the same concerns as do asbestos fibres', Report on DEFRA project CB0406
4. RCEP., 2008. Twenty-seventh Report Novel Materials in the Environment: The case of nanotechnology Royal Commission on Environmental Pollution. The Stationery Office , Norwich.
5. Stone V, Hankin S, Aitken R, Aschberger K, Baun A, Christensen F, Fernandes T, Hansen SF, Hartmann NB, Hutchinson G, Johnston H, Micheletti G, Peters S, Ross B, Sokull-Kluettgen B, Stark D, Tran L. 2009. Engineered Nanoparticles: Reviewof Health and Environmental Safety (ENRHES). Available at: http://ihcp.jrc.ec.europa.eu/whats-new/enhres-final-report (Accessed July 15, 2010)
6. NIOSH 2011. NIOSH Current Intelligence Bulletin Occupational Exposure to Carbon Nanotubes and Nanofibers. Department Of Health And Human Services Centers for Disease Control and Prevention National Institute for Occupational Safety and Health, Washington, D.C.
7. Regulation (EC) No 1272/2008 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 16 December 2008 on classification, labelling and packaging of substances and mixtures, amending and repealing Directives 67/548/EEC and 1999/45/EC, and amending Regulation (EC) No 1907/2006 (available: http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2008:353:0001:1355:EN:PDF (Accessed March 25, 2012)

APPENDIX 1: NanoRiskCat●●●|●● Template

Human hazard profile

1. **HARN: Does the nanomaterial fulfill the HARN paradigm?**

   Answer: Yes

   Arguments and explanation: Most Carbon nanotubes fulfill HARN as they have a high surface area and a length to diameter aspect ratio greater than 10 to 1. Furthermore, the diameter of the fibres is thin enough to pass ciliated airways and the length is long enough to initiate the onset of e.g. frustrated phagocytosis[9] and other inflammatory pathways; and finally, carbon nanotubes are biopersistent (Tran et al. 2008, Stone et al. 2009, NIOSH 2011).

2. **Overall evaluation of human hazard**

   We conclude that the color-code that best reflects the human hazard profile of carbon nanotubes is ● based evidence of HARN

Environment hazard profile

1. **Bulk – "Level A CLP": Is the bulk form of the nanomaterial classified as CLP Acute 1 or Chronic 1 or Chronic 2?**

   Answer: No

   Arguments and explanation: Carbon nanotubes do not have a meaningful bulk parent materials and hence the answer to this question is no by default

2. **Nano – LC_{50}<10 mg/l: Is the nanomaterial in question reported to be hazardous to environmental species i.e. LC50 or EC 50 <10 mg/l?**

   Answer: No

Arguments and explanation: To the best of our knowledge Carbon nanotubes have not been reported to be hazardous to environmental species i.e. LC50 or EC 50 <10 mg/l (Stone et al. 2009)

3. **Bulk – "Level B CLP": Is the bulk form of the nanomaterial classified as CLP Chronic 3 or Chronic 4 or documented nano-specific effects?**

Answer: No

Arguments and explanation: Carbon nanotubes do not have a meaningful bulk parent materials and hence the answer to this question is no by default

4. **Nano – LC50<100 mg/l: Is the nanomaterial in question reported to be hazardous to environmental species i.e. LC50 or EC 50 <100 mg/l?**

Answer: Yes

Arguments and explanation: Following US EPA (2002) guidelines on methods for measuring the acute toxicity of effluents and receiving waters to freshwater and marine organisms The influence of moderately hard reconstituted water containing 100 mg l^{-1} of NOM on carbon nanotubes has been studied in *Ceriodaphnia dubia* by Kennedy et al. (2008) and no significant mortality was observed after 48 hours of exposure to 32 mg l^{-1} of hydroxylated multiwalled carbon nanotubes (MWCNT–OH). After 7 days of settling, no significant effect was observed on survival after exposure to 120.2 mg l^{-1} MWCNT–OH and 88.9 mg l^{-1} MWCNT–COOH. The hydrodynamic diameter of aggregates was found to be 208.6 ± 2.2 to 223.3 ± 0.8 nm, 181.5 ± 1.1 to 187.4 ± 1.1 nm, 181.1 ± 0.3 to 185.1 ± 2.0 nm for raw MWCNT, MWCNT–OH and MWCNT–COOH, respectively. For the raw MWCNT the survival rate was down to 7±12% at a concentration of 39.5 mg l^{-1} and $LC_{50,48h}$ was determined to be 50.9 [38.4–67.6] mg l^{-1}.

5. **T½>40 days: Is the nanomaterial in question persistent i.e. T½>40 days?**

Answer: Yes

Arguments and explanation: Carbon nanotubes are often said to be some of the least biodegradable man-made materials known (RCEP 2008). Only one paper refers to the possible degradation of single-walled carbon nanotubes (SWCNT) via enzymatic catalysis (Allen *et al.* 2008). After incubation of SWCNT with a natural horseradish peroxidase (HRP) and low concentrations of H_2O_2 (40 µM) at 4°C over 12 weeks under static conditions Allen *et al.* (2008) found indications of degradation of the nanotube structure. These results lead to the suggestion that plant peroxidises may have a role in carbon nanotubes degradation along with material type and physic-chemical conditions.

6. Overall evaluation of environmental hazard

Given the fact that carbon nanotubes have been reported to be hazardous to environmental species i.e. LC50 or EC 50 <100 mg/l and T½>40 days, we conclude that the color-code that best reflects the environmental hazard profile of carbon nanotubes is ●