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***The detection of subtle changes in 3D microstructural relations in the CNS using 2nd order stereological methods measured with isotropic linear rulers.***

*Joint with Gesa Staats and Matthew Reed*

The use of isotropic linear probes to elicit 2nd order stereological estimates offers a number of advantages. As the result of a conscious effort to combine the benefits of rigorous unbiased stereological techniques with the simplest possible experimental implementation we developed the One Stop Stereology technique. Measurements can be made on thin sections with IUR or VUR sampling properties. Thus the approach can be applied to historical collections of histological material. The fact that most laboratories using stereological methods tend to sample and subsequently store material in blocks of VUR sampled tissues makes the approach potentially widely applicable. The technical aspects of the application of isotropic linear rulers has been described ([6]). The approach has been dubbed ‘One-stop stereology’ (OSS) because, at one sitting it is possible to measure: (i) the volume fraction,  $V_V(A_i, ref)$ , for each phase from the Rosiwal ([7]) relationship  $L_L = V_V$ . (ii) the surface density for each phase,  $S_V(A_i, ref)$ , from the classical relation  $S_V = 2I_L$  (e.g. [10]), (iii) the volume weighted star volume of each phase, which is equal to the volume weighted mean particle volume for a phase consisting of discrete particles, using the methods of [5, 9, 3], (iv) the contiguity and matrix of surface affinities between all pairs of phases ([4]), (v) if VUR sections are used, the ‘volume anisotropy’ using star-volumes (e.g. [2]), (vi) the volumetric set covariance,  $C(h, A_i)$  for each phase, i.e. the probability that a pair of points separated by a distance  $h$  units will simultaneously hit phase  $A_i$ . (vii) all crosscovariance functions,  $C(h, A_i, A_j)$ , for each pair of phases ( $i \neq j$ ), i.e. the probability that a pair of points separated by a distance  $h$  units will hit phases  $A_i$  and  $A_j$ , (viii) the ‘star’ covariance for each phase,  $C^*(h, A_i)$ , i.e. the probability that a pair of points separated by a distance  $h$  units will both hit phase  $A_i$  and be joined by a straight line fully within the phase ([1]), (ix) a range of linear contact distribution functions ([8]).

This paper will describe the application of OSS to the study of the effect of fetal exposure to the NMDA antagonist MK801 on the postnatal development of rat brain. Subtle changes to the spatial relationship between glial and neuronal cells in the subiculum were demonstrated through the measurement of crosscovariance. No differences between experimental and control groups were found with 1st order estimators.

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