

Supplemental Table 4: Some enzymatic dissociation methods provide reliable test results when processing of mononuclear cells from brain and spinal cord. The third pre-defined criterion of our assay qualification algorithm was test reliability. As stated in the Method section, the 95% confidence interval of the mean was calculated to express the precision and repeatability of each dissociation method tested. Then, the coefficient of variation (CV), defined as the ratio of the standard deviation to the mean was computed. The CV shows the extent of variability in relation to the mean of two experimental samples. In this study, the CV was set to $\leq 25\%$, given the heterogeneity of EAE disease activity. Following the enzymatic dissociation of brains from mice with clinical EAE, Kit, accutase 15, and papain 60 met the criteria. Kit and papain 60 were sufficiently reliable when used to dissociate spinal cords.

Brain			Mean	Standard deviation	Coefficient of Variation
Accutase 15/Papain 30	Replicate 1	Valid N: 3	2403333	891871	37.11
	Replicate 2	Valid N: 4	6300000	5175584	82.15
Kit	Replicate 1	Valid N: 3	7500000	3905125	52.07
	Replicate 2	Valid N: 4	10500000	7363876	70.13
Accutase 15	Replicate 1	Valid N: 3	793333	476480	60.06
	Replicate 2	Valid N: 5	1370000	541202	39.50
Papain 15	Replicate 1	Valid N: 4	350000	291209	83.20
	Replicate 2	Valid N: 3	956667	398037	41.61
Papain 30	Replicate 1	Valid N: 3	333667	356542	106.86
	Replicate 2	Valid N: 3	843333	549939	65.21
Papain 60	Replicate 1	Valid N: 3	3566667	2498666	70.06
	Replicate 2	Valid N: 5	534400	457479	85.61
Accutase 15/Papain 30	Replicate 1	Valid N: 3	2403333	891871	37.11
	Replicate 2	Valid N: 4	6300000	5175584	82.15
Spinal cord					
Kit	Replicate 1	Valid N: 3	1566667	1525068	97.34
	Replicate 2	Valid N: 4	1605000	1955445	121.83
Papain 60	Replicate 1	Valid N: 3	1716667	1601822	93.31
	Replicate 2	Valid N: 4	988500	811695	82.11