## High-throughput extraction and comparison of gene expression patterns from 2D RNA in situ hybridization images (Supplemental)

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## **1 SCORING METRIC COMPARISON**

Below we include the comparison of the scoring metrics for both the raw scores and RSS scores for projections along the AP axis only. Full comparisons for both AP and DV are shown in the main paper.  $\ensuremath{^*\text{to}}$  whom correspondence should be addressed

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Annotation Term	Genes	Actual				RSS			
		MSE	Haar	MI	SMI	MSE	Haar	MI	SMI
mnioserosa anlage in statu nascendi	19	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
anlage in statu nascendi	30	0.186	0.162	0.054	0.017	0.067	0.040	0.035	0.023
anterior endoderm anlage in statu nascendi	30	0.012	0.012	0.003	0.002	0.002	0.001	0.002	0.001
cellular blastoderm	27	0.140	0.105	0.355	0.267	0.088	0.105	0.429	0.338
clypeolabrum anlage in statu nascendi	10	0.011	0.007	0.010	0.005	0.040	0.041	0.004	0.007
dorsal ectoderm anlage	15	0.047	0.009	0.005	0.005	0.002	0.007	0.006	0.007
dorsal ectoderm anlage in statu nascendi	72	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
ectoderm anlage in statu nascendi	13	0.054	0.107	0.031	0.021	0.019	0.006	0.019	0.009
endoderm anlage in statu nascendi	9	0.377	0.246	0.036	0.034	0.046	0.129	0.019	0.030
foregut anlage in statu nascendi	25	0.003	0.002	0.003	0.000	0.000	0.000	0.001	0.000
gap	27	0.074	0.030	0.035	0.022	0.015	0.015	0.034	0.026
head epidermis anlage in statu nascendi	4	0.015	0.022	0.007	0.005	0.047	0.038	0.003	0.004
head epidermis dorsal anlage in statu nascendi	10	0.007	0.004	0.002	0.002	0.017	0.011	0.002	0.001
head mesoderm anlage	5	0.409	0.393	0.298	0.178	0.266	0.409	0.181	0.204
head mesoderm anlage in statu nascendi	15	0.258	0.298	0.347	0.448	0.288	0.317	0.310	0.356
hindgut anlage in statu nascendi	20	0.000	0.001	0.001	0.000	0.000	0.000	0.000	0.000
mesectoderm anlage in statu nascendi	12	0.027	0.028	0.074	0.076	0.013	0.016	0.037	0.020
mesoderm anlage in statu nascendi	18	0.334	0.496	0.331	0.205	0.128	0.292	0.221	0.195
pair rule	5	0.520	0.322	0.064	0.041	0.041	0.044	0.030	0.047
pole cell	13	0.829	0.733	0.932	0.956	0.967	0.870	0.946	0.968
posterior endoderm anlage in statu nascendi	38	0.000	0.000	0.001	0.000	0.000	0.000	0.000	0.000
procephalic ectoderm anlage in statu nascendi	67	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
segmentally repeated	9	0.001	0.001	0.002	0.001	0.000	0.000	0.002	0.001
trunk mesoderm anlage	5	0.216	0.279	0.072	0.047	0.022	0.053	0.066	0.046
trunk mesoderm anlage in statu nascendi	17	0.240	0.217	0.479	0.455	0.080	0.143	0.345	0.411
ventral ectoderm anlage	11	0.119	0.092	0.025	0.022	0.002	0.001	0.006	0.002
ventral ectoderm anlage in statu nascendi	69	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.000
visual anlage in statu nascendi	17	0.000	0.003	0.005	0.004	0.000	0.000	0.001	0.000
yolk nuclei	35	0.529	0.566	0.750	0.750	0.698	0.683	0.827	0.802

**Table 1.** Table showing the enrichment terms of only the AP projections for all genes within the second time grouping (time stages 4-6) for all four similarity values: Mean Squared Error (MSE), Haar wavelets (Haar), Mutual Information (MI), and Spatial Mutual Information (SMI) for the Actual scores, and RSS scores. Each row represents an individual annotation term, light gray shading represents 0.05 significance, while dark gray represents 0.01 significance values.