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Supplementary Material

The effect of elevated CO_2 and virus infection on the primary metabolism of wheat

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Table S1. Ion extraction window and retention times (RT) of targeted polar metabolites from standard mix (adapted from Liu and Rochfort 2013)

RT, Retention time; nd, not detected; *, amino acids essential to aphid diet according to Dadd (1985)

Compound	Ions quantified	Ion extraction window (*m/z*)	RT (min)
Amino acids			
Alanine (ala)	$[M+H]^+$	90.05-90.06	5.01
Asparagine (asn)	$[M+H]^+$	133.06-133.07	5.09
Aspartic acid (asp)	$[M+H]^+$	134.04-134.05	4.95
Arginine (arg)*	$[M+H]^+$	175.11-175.12	4.80
Glutamic acid (glu)	$[M+H]^+$	148.06-148.07	5.14
Glutamine (gln)	$[M+H]^+$	147.07-147.08	nd
Glycine (gly)	$[M+H]^+$	76.03-76.05	nd
Histidine (his)*	$[M+H]^+$	156.07-156.08	4.78
Isoleucine (iso)*	$[M+H]^+$	132.10-132.1	9.06
Leucine (leu)*	$[M+H]^+$	132.10-132.11	9.32
Lysine (lys)*	$[M+H]^+$	147.11-147.12	4.46
Methionine (met)*	$[M+H]^+$	150.05-150.06	8.42
Phenylalanine (phe)*	$[M+H]^+$	166.08-166.09	10.71
Proline (pro)	$[M+H]^+$	116.07-116.08	5.68
Serine (ser)	$[M+H]^+$	106.05-106.06	4.90
Threonine (thr)*	$[M+H]^+$	120.06-120.07	5.06
Tryptophan (trp)*	$[M+H]^+$	205.09-205.10	12.39
Tyrosine (tyr)*	$[M+H]^+$	182.08-182.09	9.09
Valine (val)*	$[M+H]^+$	118.08-118.09	6.88
Organic acids			
Succinic acid	$[M-H]^-$	116.5-117.5	nd
Citric acid	$[M-H]^-$	190.5-191.5	8.69
Malic acid	$[M-H]^-$	132.5-133.5	6.82
Tartaric acid	$[M-H]^-$	148.5-149.5	5.68
Maleic acid	$[M-H]^-$	114.5-115.5	8.74
Fumaric acid	$[M-H]^-$	114.5-115.5	9.42
trans-Aconitic acid	$[M-H]^-$	172.5-173.5	9.72
Sugars			
Mannitol	$[M+HCOO]^-$	226.5-227.5	5.22
Glucose	$[M+HCOO]^-$	224.5-225.5	5.10
Fructose	$[M+HCOO]^-$	224.5-225.5	5.27
Sucrose	$[M+HCOO]^-$	386.5-387.5	5.74
Trehalose	$[M+HCOO]^-$	386.5-387.5	5.37
Raffinose	$[M+HCOO]^-$	548.5-549.5	5.95

Table S2. Summary of ANOVA for wheat growth parameters (height, tiller number and dry weight), disease severity (% yellowing per plant) and total foliar carbon and nitrogen content

Source of variation (groups) were CO_2 by inoculation treatment. Df, degrees of freedom; SS, sum of square; MS, mean squares. Significant values are denoted by asterisks (*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$)

	df	SS	MS	F value	P value
Height					
groups	5	95.7	19.13	1.871	0.101
residuals		108	1104.6	10.23	
Tiller number					
groups	5	112.1	22.416	5.932	6.94E-05***
residuals		108	408.1	3.779	
Dry weight					
groups	5	31.29	6.258	11.89	3.50E-09***
residuals		108	56.86	0.527	
Disease severity					
groups	5	19212	3842	178.8	2.00E-16***
residuals		108	2321	21	
Carbon					
groups	5	27.28	5.455	23.41	7.18E-16***
residuals		108	25.17	0.233	
Nitrogen					
groups	5	20.25	4.049	13.56	2.91E-10***
residuals		108	32.26	299	

Table S3. The concentration of individual sugars/sugar alcohols, organic acids and amino acids (µmol/g) in uninoculated controls, sham-inoculated controls and BYDV-infected wheat (cv. Yitpi) under aCO_2 (400 µmol mol^{-1}) and eCO_2 (650 µmol mol^{-1}) (mean ± s.e.m.)

*, amino acids essential to aphid diet according to Dadd (1985)

	Uninoculated controls		Sham-inoculated controls		BYDV-infected	
	aCO_2	eCO_2	aCO_2	eCO_2	aCO_2	eCO_2
Sugars						
Trehalose	0.005 ± 0.000	0.008 ± 0.001	0.004 ± 0.000	0.007 ± 0.001	0.003 ± 0.000	0.005 ± 0.000
Mannitol	0.008 ± 0.000	0.012 ± 0.001	0.007 ± 0.000	0.011 ± 0.001	0.010 ± 0.000	0.016 ± 0.001
Raffinose	0.128 ± 0.000	0.127 ± 0.000	0.122 ± 0.000	0.136 ± 0.000	0.096 ± 0.000	0.095 ± 0.000
Sucrose	1.118 ± 0.004	1.162 ± 0.003	1.177 ± 0.009	1.165 ± 0.003	1.110 ± 0.001	1.108 ± 0.000
Fructose	1.217 ± 0.008	2.122 ± 0.010	1.192 ± 0.007	1.487 ± 0.009	1.265 ± 0.009	2.323 ± 0.000
Glucose	1.503 ± 0.013	2.769 ± 0.001	1.556 ± 0.011	1.835 ± 0.000	1.651 ± 0.015	3.036 ± 0.008
Total	*3.980 ± 0.026*	*6.300 ± 0.028*	*4.059 ± 0.024*	*4.839 ± 0.026*	*4.336 ± 0.032*	*6.584 ± 0.052*
Organic acids						
Tartaric acid	0.002 ± 0.000	0.003 ± 0.000	0.003 ± 0.000	0.002 ± 0.000	0.002 ± 0.000	0.002 ± 0.000
Fumaric acid	0.038 ± 0.003	0.032 ± 0.005	0.033 ± 0.002	0.029 ± 0.002	0.027 ± 0.003	0.023 ± 0.003
Citric acid	0.815 ± 0.005	0.864 ± 0.005	0.804 ± 0.000	0.862 ± 0.008	0.630 ± 0.005	0.581 ± 0.008
trans-Aconitic acid	3.546 ± 0.006	3.964 ± 0.007	4.214 ± 0.001	3.838 ± 0.006	3.727 ± 0.009	3.459 ± 0.005
Total	*4.402 ± 0.040*	*4.863 ± 0.008*	*5.061 ± 0.004*	*4.731 ± 0.006*	*4.382 ± 0.002*	*4.066 ± 0.006*
Amino acids						
Histidine*	0.034 ± 0.004	0.046 ± 0.008	0.036 ± 0.005	0.059 ± 0.007	0.110 ± 0.006	0.082 ± 0.001
Arginine*	0.364 ± 0.022	0.368 ± 0.003	0.292 ± 0.015	0.338 ± 0.005	0.319 ± 0.003	0.344 ± 0.007

Phenylalanine*	0.898	±	0.09 0	0.736	±	0.06 4	0.785	±	0.03 3	0.796	±	0.05 9	1.180	±	0.16 0	1.032	±	0.17 1
Tyrosine*	0.555	±	0.04 1	0.456	±	0.03 5	0.525	±	0.02 3	0.510	±	0.03 7	0.578	±	0.05 9	0.548	±	0.08 1
Isoleucine*	0.888	±	0.07 0	0.711	±	0.06 4	0.772	±	0.03 6	0.825	±	0.07 1	0.888	±	0.09 1	0.818	±	0.11 3
Tryptophan*	0.614	±	0.05 3	0.631	±	0.05 7	0.434	±	0.02 5	0.723	±	0.05 7	1.654	±	0.20 1	1.540	±	0.25 6
Leucine*	1.506	±	0.11 4	1.202	±	0.10 8	1.320	±	0.06 2	1.335	±	0.11 3	1.345	±	0.12 0	1.297	±	0.17 2
Lysine*	1.211	±	0.08 3	1.303	±	0.11 1	1.114	±	0.06 2	1.296	±	0.09 5	1.301	±	0.10 8	1.511	±	0.19 0
Threonine*	1.656	±	0.09 5	1.636	±	0.09 5	1.898	±	0.08 9	1.684	±	0.08 4	1.934	±	0.19 2	1.649	±	0.19 9
Valine*	1.790	±	0.21 1	1.527	±	0.13 2	2.213	±	0.15 9	1.858	±	0.15 4	2.356	±	0.22 5	1.924	±	0.32 8
Proline	0.935	±	0.37 5	0.746	±	0.17 7	0.636	±	0.05 1	0.657	±	0.08 5	1.315	±	0.33 3	1.030	±	0.16 3
Alanine	1.574	±	0.07 0	1.364	±	0.07 7	1.564	±	0.06 2	1.347	±	0.05 7	1.674	±	0.18 5	1.202	±	0.13 8
Serine	3.551	±	0.09 8	3.405	±	0.15 4	3.599	±	0.14 0	3.334	±	0.09 1	3.990	±	0.31 8	3.194	±	0.33 5
Aspartic acid	4.364	±	0.17 1	3.784	±	0.31 1	3.669	±	0.14 8	3.964	±	0.21 8	3.292	±	0.28 2	2.943	±	0.27 7
Glutamic acid	10.300	±	0.65 8	9.878	±	0.58 1	13.243	±	0.63 3	11.915	±	0.65 1	10.394	±	0.91 3	9.383	±	1.09 1
Total	*30.241*	±	*0.928*	*27.794*	±	*1.046*	*32.100*	±	*4.863*	*30.640*	±	*0.960*	*32.329*	±	*1.512*	*28.499*	±	*1.703*