**Data**

**Title of MS:** Biosynthesis of antioxidative enzymes and polyphenolics content in calli cultures of *Prunella vulgaris* L. in response to auxins and cytokinins

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**MS number:** LABB-2019-1993.

**Data of Figure 3a:** Effect of full strength MS medium with different concentrations of NAA on growth kinetics and fresh biomass production in *Prunella vulgaris*. Data was collected for & 7 weeks with 7 days intervals. From three replicates, values are the mean ± standard error.

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| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Days** | **NAA 0.5** **(mg l-1)** | **SE (±)** | **NAA 1.0****(mg l-1)** | **SE (±)** | **NAA 1.5 (mg l-1)** | **SE (±)** | **NAA 2.0 (mg l-1)** | **SE (±)** |
| 1 | 0.0043 | 0.00011 | 0.01 | 0.001 | 0.0043 | 0.001 | 0.007 | 0.001 |
| 07 | 0.4023 | 0.02011 | 0.3885 | 0.02011 | 0.399 | 0.01943 | 0.423 | 0.02115 |
| 14 | 0.49 | 0.0245 | 0.4425 | 0.0245 | 0.418 | 0.02212 | 0.495 | 0.02475 |
| 21 | 0.54 | 0.027 | 0.443 | 0.027 | 0.42 | 0.02215 | 0.514 | 0.0257 |
| 28 | 0.715 | 0.03575 | 0.488 | 0.03575 | 0.47 | 0.0244 | 0.59 | 0.0295 |
| 35 | 0.718 | 0.0359 | 0.8035 | 0.0359 | 0.65 | 0.04018 | 0.61 | 0.0305 |
| 42 | 0.73 | 0.0365 | 0.553 | 0.0365 | 0.624 | 0.02765 | 0.477 | 0.02385 |
| 49 | 0.563 | 0.02815 | 0.62 | 0.02815 | 0.449 | 0.031 | 0.75 | 0.0375 |

**Data of Figure 3b:** Effect of half strength MS medium with different concentrations of NAA on growth kinetics and fresh biomass production in *Prunella vulgaris*. Data was collected for & 7 weeks with 7 days intervals. From three replicates, values are the mean ± standard error.

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| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Days** | **HS-MS-NAA 0.5** **(mg l-1)** | **SE (±)** | **HS-MS-NAA 1.0****(mg l-1)** | **SE (±)** | **HS-MS-NAA 1.5 (mg l-1)** | **SE (±)** | **HS-MS-NAA 2.0 (mg l-1)** | **SE (±)** |
| 1 | 0.007 | 0.001 | 0.007 | 0.001 | 0.007 | 0.001 | 0.007 | 0.001 |
| 07 | 0.433 | 0.02075 | 0.44 | 0.02165 | 0.421 | 0.00842 | 0.415 | 0.0176 |
| 14 | 0.492 | 0.0215 | 0.48 | 0.0246 | 0.44 | 0.0088 | 0.43 | 0.0192 |
| 21 | 0.51 | 0.024 | 0.52 | 0.0255 | 0.503 | 0.01006 | 0.48 | 0.0208 |
| 28 | 0.563 | 0.0242 | 0.443 | 0.02815 | 0.55 | 0.011 | 0.484 | 0.01772 |
| 35 | 0.56 | 0.031 | 0.563 | 0.028 | 0.58 | 0.0116 | 0.62 | 0.02252 |
| 42 | 0.76 | 0.03565 | 0.69 | 0.038 | 0.67 | 0.0134 | 0.713 | 0.0276 |
| 49 | 0.539 | 0.0235 | 0.495 | 0.02695 | 0.497 | 0.00994 | 0.47 | 0.0198 |

*HS-MS (Half strength Murashige and Skoog medium)*

**Data of Figure 3c:** Effect of different concentrations of NAA with BA on growth kinetics and fresh biomass production in *Prunella vulgaris*. Data was collected for & 7 weeks with 7 days intervals. From three replicates, values are the mean ± standard error.

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| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Days** | **NAA+BA 0.5****(mg l-1)** | **SE (±)** | **NAA+BA 1.0****(mg l-1)** | **SE (±)** | **NAA+BA 1.5 (mg l-1)** | **SE (±)** | **NAA+BA 2.0 (mg l-1)** | **SE (±)** |
| 1 | 0.0043 | 0.001 | 0.023 | 0.00115 | 0.0043 | 0.001 | 0.09 | 0.0045 |
| 07 | 0.461 | 0.02305 | 0.44 | 0.022 | 0.472 | 0.01416 | 0.31 | 0.0155 |
| 14 | 0.443 | 0.02215 | 0.49 | 0.0245 | 0.485 | 0.01455 | 0.325 | 0.01625 |
| 21 | 0.542 | 0.0271 | 0.49 | 0.0245 | 0.531 | 0.01593 | 0.35 | 0.0175 |
| 28 | 0.472 | 0.0236 | 0.67 | 0.0335 | 0.496 | 0.01488 | 0.399 | 0.01995 |
| 35 | 0.609 | 0.03045 | 0.741 | 0.03705 | 0.642 | 0.01926 | 0.57 | 0.0285 |
| 42 | 0.75 | 0.0375 | 0.95 | 0.0475 | 0.74 | 0.0222 | 0.87 | 0.0435 |
| 49 | 0.56 | 0.028 | 0.394 | 0.0197 | 0.53 | 0.0159 | 0.35 | 0.0175 |

**Data of Figure 3d:** Effect of different concentrations of NAA with 2, 4-D on growth kinetics and fresh biomass production *Prunella vulgaris*. Data was collected for & 7 weeks with 7 days intervals. From three replicates, values are the mean ± standard error.

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| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Days** | **0.5 NAA+1.0 2, 4-D** **(mg l-1)** | **SE (±)** | **0.5 NAA+2.0 2, 4-D** **(mg l-1)** | **SE (±)** | **1.0 NAA+0.5 2, 4-D** **(mg l-1)** | **SE (±)** | **NAA+2, 4-D (1.0)****(mg l-1)** | **SE (±)** | **2.0 NAA+ 0.5 2, 4-D****(mg l-1)** | **SE (±)** | **NAA+ 2, 4-D (2.0)****(mg l-1)** | **SE (±)** |
| 1 | 0.006 | 0.001 | 0.006 | 0.001 | 0.008 | 0.001 | 0.023 | 0.00115 | 0.008 | 0.001 | 0.15 | 0.0045 |
| 07 | 0.31 | 0.0155 | 0.25 | 0.0125 | 0.29 | 0.0145 | 0.332 | 0.0166 | 0.314 | 0.0157 | 0.573 | 0.01719 |
| 14 | 0.501 | 0.02505 | 0.323 | 0.01615 | 0.35 | 0.0175 | 0.343 | 0.01715 | 0.3714 | 0.01857 | 0.761 | 0.02283 |
| 21 | 0.61 | 0.0305 | 0.34 | 0.017 | 0.37 | 0.0185 | 0.38 | 0.019 | 0.244 | 0.0122 | 0.999 | 0.02997 |
| 28 | 0.71 | 0.0355 | 0.68 | 0.034 | 0.943 | 0.04715 | 0.96 | 0.048 | 0.55 | 0.0275 | 1.3 | 0.039 |
| 35 | 0.35 | 0.0175 | 0.38 | 0.019 | 0.65 | 0.0325 | 0.5 | 0.025 | 0.416 | 0.0208 | 1.03 | 0.0309 |
| 42 | 0.405 | 0.02025 | 0.47 | 0.0235 | 0.39 | 0.0195 | 0.412 | 0.0206 | 0.402 | 0.0201 | 0.764 | 0.02292 |
| 49 | 0.42 | 0.021 | 0.47 | 0.0235 | 0.37 | 0.0185 | 0.34 | 0.017 | 0.46 | 0.023 | 0.65 | 0.0195 |

**Data of Figure 3e:** Effects of different concentrations 2, 4-D alone or in combination with BA, or with BA and NAA or 2, 4-D in combination with GA3 on growth kinetics, and fresh biomass production *Prunella vulgaris*. Data was collected for & 7 weeks with 7 days intervals. From three replicates, values are the mean ± standard error.

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| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Days** | **2, 4-D (1.0)****(mg l-1)** | **SE (±)** | **2, 4-D (2.0)****(mg l-1)** | **SE (±)** | **BA+2, 4-D (1.0) (mg l-1)** | **SE (±)** | **BA+NAA+2, 4-D (0.5) (mg l-1)** | **SE (±)** | **GA3+ 2, 4-D (1.0) (mg l-1)** | **SE (±)** | **GA3+ 2, 4-D (2.0) (mg l-1)** | **SE (±)** |
| 1 | 0.004 | 0.001 | 0.004 | 0.001 | 0.0032 | 0.001 | 0.124 | 0.0062 | 0.01 | 0.001 | 0.021 | 0.00105 |
| 7 | 0.31 | 0.015 | 0.274 | 0.013 | 0.299 | 0.014 | 0.297 | 0.01485 | 0.285 | 0.01425 | 0.284 | 0.0142 |
| 14 | 0.442 | 0.022 | 0.332 | 0.016 | 0.35 | 0.017 | 0.293 | 0.01465 | 0.32 | 0.016 | 0.27 | 0.0135 |
| 21 | 0.46 | 0.023 | 0.42 | 0.021 | 0.382 | 0.019 | 0.306 | 0.0153 | 0.373 | 0.01865 | 0.473 | 0.02365 |
| 28 | 0.65 | 0.032 | 0.67 | 0.033 | 0.389 | 0.019 | 0.641 | 0.03205 | 0.57 | 0.0285 | 0.785 | 0.03925 |
| 35 | 0.86 | 0.043 | 0.95 | 0.047 | 0.78 | 0.039 | 1.03 | 0.0515 | 0.587 | 0.02935 | 0.85 | 0.0425 |
| 42 | 0.49 | 0.024 | 0.309 | 0.015 | 0.41 | 0.020 | 0.252 | 0.0126 | 0.45 | 0.0225 | 0.43 | 0.0215 |
| 49 | 0.357 | 0.017 | 0.254 | 0.013 | 0.382 | 0.019 | 0.382 | 0.0191 | 0.26 | 0.013 | 0.369 | 0.01845 |

**Data of Figure 4:** Correlation of fresh biomass production with superoxide dismutase and peroxidase activities in calli cultures of *Prunella vulgaris*. From three replicates, values are the mean ± standard error.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Treatments** | **Fresh weight** | **POD** | **SE (±)** | **SOD** | **SE (±)** |
| T1 | 1.357 | 0.06785 | 0.01227 | 0.31111 | 0.00933 |
| T2 | 1.843 | 0.09215 | 0.00141 | 0.13552 | 0.00407 |
| T3 | 4.032 | 0.2016 | 0.00194 | 0.04914 | 0.00147 |
| T4 | 0.676 | 0.0338 | 0.00144 | 0.1687 | 0.00506 |
| T5 | 0.62 | 0.031 | 0.0049 | 0.30516 | 0.00915 |
| T6 | 0.9982 | 0.04991 | 0.00647 | 0.21628 | 0.00649 |
| T7 | 5.909 | 0.29545 | 0.00329 | 0.12144 | 0.00364 |
| T8 | 0.1054 | 0.00527 | 0.01241 | 0.18748 | 0.00562 |
| T9 | 5.231 | 0.26155 | 0.00781 | 0.32269 | 0.00968 |
| T10 | 0.331 | 0.01655 | 0.01053 | 0.19656 | 0.0059 |
| T11 | 5.215 | 0.26075 | 0.01543 | 0.08607 | 0.00258 |
| T12 | 6.325 | 0.31625 | 0.00235 | 0.21346 | 0.0064 |
| T13 | 2.048 | 0.1024 | 0.00168 | 0.15806 | 0.00474 |
| T14 | 0.0663 | 0.00332 | 0.00496 | 0.17402 | 0.00522 |
| T15 | 0.3441 | 0.01721 | 0.00334 | 0.36651 | 0.011 |
| T16 | 0.172 | 0.0086 | 0.00133 | 0.11393 | 0.00342 |
| T17 | 0.558 | 0.0279 | 0.00516 | 0.2385 | 0.00715 |
| T18 | 0.12 | 0.006 | 0.00721 | 0.31925 | 0.00958 |
| T19 | 1.166 | 0.0583 | 0.00345 | 0.21596 | 0.00648 |
| T20 | 0.365 | 0.01825 | 0.00121 | 0.22222 | 0.00667 |
| T21 | 0.52 | 0.026 | 0.0104 | 0.34773 | 0.01043 |
| T22 | 0.738 | 0.0369 | 0.00801 | 0.12676 | 0.0038 |
| T23 | 0.314 | 0.0157 | 0.00161 | 0.19499 | 0.00585 |
| T24 | 2.938 | 0.1469 | 0.01526 | 0.12175 | 0.00365 |
| Control | -- | -- | -- | -- | -- |

**Data of Figure 5:** Correlation of dry biomass accumulation with TPC and TFC in calli cultures of *Prunella vulgaris*. From three replicates, values are the mean ± standard error.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Treatments** | **Dry weight** | **SE (±)** | **TPC** | **SE (±)** | **TFC** | **SE (±)** |
| T1 | 0.1964 | 0.00982 | 6.42939 | 0.32147 | 13.52222 | 0.67611 |
| T2 | 0.297 | 0.01485 | 8.86847 | 0.44342 | 16.97594 | 0.8488 |
| T3 | 0.2932 | 0.01466 | 7.92211 | 0.39611 | 15.2003 | 0.76002 |
| T4 | 0.109 | 0.00545 | 4.22447 | 0.21122 | 6.90745 | 0.34537 |
| T5 | 0.1 | 0.005 | 15.18078 | 0.75904 | 14.74175 | 0.73709 |
| T6 | 0.161 | 0.00805 | 9.86361 | 0.49318 | 8.40992 | 0.4205 |
| T7 | 0.4367 | 0.02184 | 14.38077 | 0.71904 | 6.77086 | 0.33854 |
| T8 | 0.017 | 0.011 | 12.33 | 0.6165 | 7.88443 | 0.39422 |
| T9 | 0.7541 | 0.03771 | 13.99052 | 0.69953 | 15.40518 | 0.77026 |
| T10 | 0.102 | 0.0051 | 6.50939 | 0.32547 | 6.93672 | 0.34684 |
| T11 | 0.4869 | 0.02434 | 9.73677 | 0.48684 | 18.05889 | 0.90294 |
| T12 | 0.4859 | 0.0243 | 18.83939 | 0.94197 | 13.62953 | 0.68148 |
| T13 | 0.2278 | 0.01139 | 7.61966 | 0.38098 | 16.49788 | 0.82489 |
| T14 | 0.0107 | 0.001 | 3.07323 | 0.15366 | 0.95612 | 0.04781 |
| T15 | 0.0555 | 0.00278 | 1.64881 | 0.08244 | 3.77568 | 0.18878 |
| T16 | 0.0538 | 0.00269 | 3.66836 | 0.18342 | 9.05383 | 0.45269 |
| T17 | 0.09 | 0.0045 | 3.77568 | 0.18878 | 8.63432 | 0.43172 |
| T18 | 0.0194 | 0.001 | 1.58052 | 0.07903 | 2.46834 | 0.12342 |
| T19 | 0.1297 | 0.00649 | 2.4098 | 0.12049 | 9.79531 | 0.48977 |
| T20 | 0.048 | 0.0024 | 0.25366 | 0.01268 | 4.07813 | 0.20391 |
| T21 | 0.0577 | 0.00289 | 3.66836 | 0.18342 | 5.61962 | 0.28098 |
| T22 | 0.0849 | 0.00424 | 5.06351 | 0.25318 | 8.47821 | 0.42391 |
| T23 | 0.0443 | 0.00222 | 3.17079 | 0.15854 | 4.89766 | 0.24488 |
| T24 | 0.2263 | 0.01131 | 8.87822 | 0.44391 | 8.55626 | 0.42781 |
| Control | -- | -- | -- | -- | -- | -- |

**Data of Figure 6:** Effects of PGRs on total protein content in callus cultures of *Prunella vulgaris*. From three replicates, values are the mean ± standard error.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatments** | **Fresh weight** | **SE (±)** | **Total protein content** | **SE (±)** |
| T1 | 1.357 | 0.06785 | 234 | 7.02 |
| T2 | 1.843 | 0.09215 | 240 | 7.2 |
| T3 | 4.032 | 0.2016 | 716 | 21.48 |
| T4 | 0.676 | 0.0338 | 186 | 5.58 |
| T5 | 0.62 | 0.031 | 834 | 25.02 |
| T6 | 0.9982 | 0.04991 | 735 | 22.05 |
| T7 | 5.909 | 0.29545 | 278 | 8.34 |
| T8 | 0.1054 | 0.00527 | 290 | 8.7 |
| T9 | 5.231 | 0.26155 | 441 | 13.23 |
| T10 | 0.331 | 0.01655 | 195 | 5.85 |
| T11 | 5.215 | 0.26075 | 492 | 14.76 |
| T12 | 6.325 | 0.31625 | 172 | 5.16 |
| T13 | 2.048 | 0.1024 | 139 | 4.17 |
| T14 | 0.0663 | 0.09331 | 290 | 8.7 |
| T15 | 0.3441 | 0.01721 | 442 | 13.26 |
| T16 | 0.172 | 0.0086 | 119 | 3.57 |
| T17 | 0.558 | 0.10279 | 353 | 10.59 |
| T18 | 0.12 | 0.26 | 336 | 10.08 |
| T19 | 1.166 | 0.10583 | 138 | 4.14 |
| T20 | 0.365 | 0.01825 | 225 | 6.75 |
| T21 | 0.52 | 0.1026 | 520 | 15.6 |
| T22 | 0.738 | 0.0369 | 337 | 10.11 |
| T23 | 0.314 | 0.0157 | 338 | 10.14 |
| T24 | 2.938 | 0.1469 | 405 | 12.15 |
| Control | -- | -- | -- | -- |

**Data of Figure 7:** Effects PGRs on antioxidant activity in callus cultures of *Prunella vulgaris*. From three replicates values are the mean ± standard error.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Treatments** | **Antioxidant activity (DRSA %)** | **SE (±)** | **Dry weight** | **SE (±)** |
| T1 | 55.89 | 2.7945 | 0.1964 | 0.00982 |
| T2 | 57.45 | 2.8725 | 0.297 | 0.01485 |
| T3 | 60.41 | 3.0205 | 0.2932 | 0.01466 |
| T4 | 56.44 | 2.822 | 0.109 | 0.00545 |
| T5 | 77.81 | 3.8905 | 0.1 | 0.005 |
| T6 | 87.4 | 4.37 | 0.161 | 0.00805 |
| T7 | 69.59 | 3.4795 | 0.4367 | 0.02184 |
| T8 | 60.22 | 3.011 | 0.017 | 0.05 |
| T9 | 60 | 3 | 0.7541 | 0.03771 |
| T10 | 52.19 | 2.6095 | 0.102 | 0.0051 |
| T11 | 57.81 | 2.8905 | 0.4869 | 0.02434 |
| T12 | 52.19 | 2.6095 | 0.4859 | 0.0243 |
| T13 | 62.05 | 3.1025 | 0.2278 | 0.01139 |
| T14 | 62.05 | 3.1025 | 0.0107 | 0.03 |
| T15 | 57.95 | 2.8975 | 0.0555 | 0.00278 |
| T16 | 66.16 | 3.308 | 0.0538 | 0.00269 |
| T17 | 56.3 | 2.815 | 0.09 | 0.0045 |
| T18 | 68.36 | 3.418 | 0.0194 | 0.03 |
| T19 | 62.6 | 3.13 | 0.1297 | 0.00649 |
| T20 | 62.4 | 3.12 | 0.048 | 0.0024 |
| T21 | 77.53 | 3.8765 | 0.0577 | 0.00289 |
| T22 | 66.03 | 3.3015 | 0.0849 | 0.00424 |
| T23 | 55.34 | 2.767 | 0.0443 | 0.00222 |
| T24 | 55.66 | 2.783 | 0.2263 | 0.01131 |
| Control | -- | -- | -- | -- |