Moisture effects on microbial protein biosynthesis from ammonium and nitrate in an unfertilised grassland

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**Figure S1:** Total and individual hydrolysable amino acid concentrations in soil following addition of 15NH4NO3 or NH415NO3 at differing WFPS over 10 days. All values are mean ± SE (*n*=3).

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**Table S1:** Summary of soil properties before N application (Loick et al., 2021). Values are mean ± SE (*n*=3).

|  |  |
| --- | --- |
| Parameter | Mean (±SE) |
| pH (water 1:2.5 *w*/*v*) | 5.6 ± 0.27 |
| Bulk density (g cm-2) | 0.8 ± 0.0005 |
| Total N (g kg−1 dry soil) | 5.0 ± 0.10 |
| Total extractable oxidized nitrogen (mg kg−1 dry soil) | 15.1 ± 0.09 |
| Ammonium N (mg kg−1 dry soil) | 9.2 ± 0.09 |
| THAA (g N kg−1 dry soil) | 1.7 ± 0.4 |
| Total organic carbon (% *w*/*w*) | * 1. 0.17 |

**Table S2:** Results of mixed effect models for amino acid concentrations over 10 d, following application of 15NH4NO3 or NH415NO3 (*n*=6). Values in bold indicate there was a significant difference (*p*<0.05).

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| AA | Time | | WFPS | | Time\*WFPS | |
| F(7,95) | *p* | F(2,950 | *p* | F(9,95) | *p* |
| Ala | **5.123** | **<0.001** | **13.543** | **<0.001** | **10.379** | **<0.001** |
| Asx | 1.745 | 0.108 | **15.642** | **<0.001** | **3.960** | **<0.001** |
| Glx | 1.500 | 0.177 | **24.745** | **<0.001** | **4.357** | **<0.001** |
| Gly | **6.922** | **<0.001** | **26.814** | **<0.001** | **18.028** | **<0.001** |
| Hyp | **7.026** | **<0.001** | **24.896** | **<0.001** | **10.689** | **<0.001** |
| Leu | **15.559** | **<0.001** | **30.480** | **<0.001** | **26.654** | **<0.001** |
| Lys | **4.664** | **<0.001** | **9.916** | **<0.001** | **7.004** | **<0.001** |
| Phe | **10.394** | **<0.001** | **10.889** | **<0.001** | **6.796** | **<0.001** |
| Pro | **4.924** | **<0.001** | **7.563** | **<0.001** | **8.829** | **<0.001** |
| Ser | 1.922 | 0.074 | **5.171** | **0.007** | **7.496** | **<0.001** |
| Thr | 0.436 | 0.877 | **4.865** | **0.010** | **4.832** | **<0.001** |
| Tyr | **3.275** | **0.004** | **7.927** | **<0.001** | **5.521** | **<0.001** |
| Val | **2.255** | **0.036** | **3.449** | **0.036** | **6.691** | **<0.001** |
| THAA |  |  |  |  |  |  |

**Table S3:** Assimilation rates into individual and total (THAAs) amino acids over 10 d following application of 15NH4NO3, NH415NO3, or the combination of these treatments. The rate units are all % day‑1. Poor fits are highlighted in red.

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | 55% | | | | | | 70% | | | | | | 85% | | | | | |
|  | 15NH4NO3 | | NH415NO3 | | Combined | | 15NH4NO3 | | NH415NO3 | | Combined | | 15NH4NO3 | | NH415NO3 | | Combined | |
| AA | Rate | *r2* | Rate | *r2* | Rate | *r2* | Rate | *r2* | Rate | *r2* | Rate | *r2* | Rate | *r2* | Rate | *r2* | Rate | *r2* |
| Ala | 0.040 | 0.69 | 0.001 | 0.19 | 0.031 | 0.44 | 0.022 | 0.71 | 0.024 | 0.49 | 0.012 | 0.05 | 0.053 | 0.93 | 0.025 | 0.39 | 0.030 | 0.48 |
| Gly | 0.033 | 0.67 | 0.004 | 0.73 | 0.028 | 0.58 | 0.008 | 0.46 | 0.008 | 0.40 | 0.018 | 0.24 | 0.046 | 0.88 | 0.010 | 0.44 | 0.012 | 0.40 |
| Val | 0.006 | 0.60 | -0.002 | 0.42 | 0.005 | 0.50 | 0.001 | 0.20 | 0.002 | 0.22 | 0.002 | 0.07 | 0.008 | 0.75 | 0.0006 | 0.08 | 0.001 | 0.08 |
| leu | 0.010 | 0.79 | 0.0001 | 0.10 | 0.006 | 0.33 | 0.006 | 0.80 | 0.002 | 0.30 | 0.011 | 0.63 | 0.016 | 0.97 | 0.0005 | 0.62 | 0.002 | 0.51 |
| Thr | 0.006 | 0.38 | -0.006 | 0.74 | 0.014 | 0.36 | 0.005 | 0.26 | 0.005 | 0.21 | -0.054 | 0.28 | 0.015 | 0.85 | 0.033 | 0.47 | 0.036 | 0.52 |
| Ser | 0.005 | 0.39 | 0.042 | 0.30 | 0.006 | 0.37 | 0.004 | 0.53 | 0.006 | 0.17 | -0.024 | 0.16 | 0.015 | 0.68 | 0.014 | 0.60 | 0.016 | 0.71 |
| Pro | 0.009 | 0.29 | 0.0003 | 0.46 | 0.006 | 0.14 | 0.008 | 0.74 | 0.004 | 0.32 | 0.003 | 0.02 | 0.021 | 0.92 | 0.017 | 0.50 | 0.014 | 0.62 |
| Asx | 0.018 | 0.77 | 0.0003 | 0.02 | 0.018 | 0.79 | 0.003 | 0.29 | 0.0002 | 0.09 | 0.003 | 0.09 | 0.015 | 0.84 | 0.0005 | 0.02 | 0.001 | 0.08 |
| Glx | 0.045 | 0.82 | 0.0009 | 0.50 | 0.038 | 0.73 | 0.009 | 0.37 | 0.003 | 0.13 | 0.039 | 0.74 | 0.045 | 0.91 | 0.0008 | 0.25 | -0.002 | 0.03 |
| Hyp | 0.0002 | 0.13 | 0.0001 | 0.27 | 0.000 | 0.04 | 0.000 | 0.21 | 0.001 | 0.44 | -0.001 | 0.24 | 0.0007 | 0.58 | 0.001 | 0.35 | 0.001 | 0.34 |
| Phe | 0.002 | 0.69 | 0.000 | 0.53 | -0.006 | 0.19 | 0.012 | 0.70 | 0.0006 | 0.31 | 0.000 | 0.00 | 0.003 | 0.91 | 0.0002 | 0.16 | 0.006 | 0.67 |
| Lys | 0.011 | 0.73 | 0.001 | 0.10 | 0.015 | 0.68 | 0.0003 | 0.00 | -0.004 | 0.12 | 0.011 | 0.38 | 0.017 | 0.82 | 0.0002 | 0.16 | -0.005 | 0.31 |
| Tyr | 0.0002 | 0.26 | 0.000 | 0.10 | 0.000 | 0.00 | 0.0002 | 0.68 | 0.0001 | 0.26 | 0.001 | 0.80 | 0.0007 | 0.84 | 0.000 | 0.18 | 0.000 | 0.21 |
| THAA | 0.184 | 0.79 | 0.010 | 0.66 | 0.161 | 0.69 | 0.078 | 0.68 | 0.052 | 0.27 | 0.020 | 0.01 | 0.25 | 0.95 | 0.096 | 0.72 | 0.113 | 0.70 |

**Table S4:** Incorporation (%I) into individual and total (THAAs) amino acids of applied 15NH4NO3, NH415NO3, or the combination of these treatments at 10 d at differing WFPS. The units for %I and SE are % and all values are mean (*n*=3).

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | 55% | | | | | | 70% | | | | | | 85% | | | | | |
|  | 15NH4NO3 | | NH415NO3 | | Combined | | 15NH4NO3 | | NH415NO3 | | Combined | | 15NH4NO3 | | NH415NO3 | | Combined | |
| AA | %I | SE | %I | SE | %I | SE | %I | SE | %I | SE | %I | SE | %I | SE | %I | SE | %I | SE |
| Ala | 0.34 | 0.05 | 0.21 | 0.06 | 0.55 | 0.07 | 0.25 | 0.03 | 0.34 | 0.07 | 0.59 | 0.06 | 0.51 | 0.04 | 0.25 | 0.14 | 0.76 | 0.18 |
| Gly | 0.24 | 0.04 | 0.10 | 0.01 | 0.34 | 0.04 | 0.09 | 0.01 | 0.11 | 0.02 | 0.20 | 0.02 | 0.42 | 0.07 | 0.14 | 0.04 | 0.56 | 0.12 |
| Val | 0.06 | 0.02 | 0.01 | 0.006 | 0.07 | 0.02 | 0.02 | 0.003 | 0.03 | 0.01 | 0.05 | 0.01 | 0.07 | 0.02 | 0.01 | 0.006 | 0.08 | 0.02 |
| Leu | 0.10 | 0.03 | 0.005 | 0.002 | 0.10 | 0.03 | 0.06 | 0.01 | 0.02 | 0.004 | 0.08 | 0.01 | 0.15 | 0.01 | 0.005 | 0.001 | 0.15 | 0.008 |
| Thr | 0.05 | 0.02 | 0.22 | 0.06 | 0.27 | 0.05 | 0.05 | 0.01 | 0.24 | 0.08 | 0.29 | 0.07 | 0.14 | 0.02 | 0.44 | 0.09 | 0.58 | 0.08 |
| Ser | 0.04 | 0.01 | 0.37 | 0.02 | 0.41 | 0.02 | 0.03 | 0.005 | 0.14 | 0.05 | 0.17 | 0.05 | 0.14 | 0.05 | 0.17 | 0.02 | 0.32 | 0.04 |
| Pro | 0.06 | 0.02 | 0.03 | 0.005 | 0.09 | 0.03 | 0.09 | 0.02 | 0.08 | 0.01 | 0.16 | 0.02 | 0.19 | 0.02 | 0.12 | 0.05 | 0.31 | 0.07 |
| Asx | 0.16 | 0.04 | 0.04 | 0.006 | 0.19 | 0.04 | 0.03 | 0.00 | 0.04 | 0.01 | 0.07 | 0.004 | 0.14 | 0.03 | 0.03 | 0.005 | 0.17 | 0.03 |
| Glx | 0.44 | 0.09 | 0.02 | 0.003 | 0.46 | 0.09 | 0.13 | 0.006 | 0.02 | 0.003 | 0.14 | 0.01 | 0.44 | 0.05 | 0.02 | 0.005 | 0.46 | 0.05 |
| Hyp | 0.002 | 0.002 | 0.01 | 0.001 | 0.01 | 0.002 | 0.003 | 0.002 | 0.02 | 0.004 | 0.02 | 0.004 | 0.01 | 0.002 | 0.01 | 0.006 | 0.02 | 0.004 |
| Phe | 0.02 | 0.005 | 0.01 | 0.002 | 0.03 | 0.005 | 0.13 | 0.03 | 0.01 | 0.002 | 0.13 | 0.03 | 0.03 | 0.002 | 0.01 | 0.000 | 0.04 | 0.002 |
| Lys | 0.11 | 0.02 | 0.03 | 0.006 | 0.14 | 0.02 | 0.04 | 0.006 | 0.002 | 0.001 | 0.04 | 0.006 | 0.15 | 0.04 | 0.01 | 0.001 | 0.16 | 0.04 |
| Tyr | 0.003 | 0.001 | 0.000 | 0.000 | 0.003 | 0.001 | 0.002 | 0.000 | 0.001 | 0.001 | 0.004 | 0.001 | 0.01 | 0.000 | 0.000 | 0.000 | 0.007 | 0.000 |
| THAA | 1.61 | 0.30 | 0.71 | 0.11 | 2.32 | 0.20 | 0.92 | 0.12 | 1.04 | 0.15 | 1.96 | 0.06 | 2.39 | 0.17 | 1.22 | 0.16 | 3.61 | 0.26 |