

# **Supplementary Materials: Occurrence of 26 Mycotoxins in Grain of Cereals Cultivated in Poland**

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**Table S1.** Limits of quantification (LOQ) ( $\mu\text{g}\cdot\text{kg}^{-1}$ ) and coefficients of correlation ( $r$ ) obtained for wheat, triticale, barley and oats.

| Mycotoxin        | Precursor Ion (m/z)                                                       | Retention Time $t_r$ (min) | Internal Standard                          | Wheat |        | Triticale |        | Barley |        | Oats |        |
|------------------|---------------------------------------------------------------------------|----------------------------|--------------------------------------------|-------|--------|-----------|--------|--------|--------|------|--------|
|                  |                                                                           |                            |                                            | LOQ   | $r$    | LOQ       | $r$    | LOQ    | $r$    | LOQ  | $r$    |
| DON              | 319.02 [M + Na] <sup>+</sup> , 297.08 [M + H] <sup>+</sup>                | 3.72                       | <sup>13</sup> C-DAS                        | 25    | 0.9923 | 25        | 0.9821 | 40     | 0.9823 | 40   | 0.9911 |
| DON-3G           | 481.15 [M + Na] <sup>+</sup>                                              | 3.94                       | <sup>13</sup> C-DAS                        | 30    | 0.9917 | 30        | 0.9931 | 40     | 0.9816 | 40   | 0.9963 |
| FUS-X            | 377.12 [M + Na] <sup>+</sup> , 372.13 [M + NH <sub>4</sub> ] <sup>+</sup> | 4.91                       | <sup>13</sup> C-DAS                        | 13    | 0.9905 | 13        | 0.9899 | 20     | 0.9929 | 20   | 0.9819 |
| NEO              | 405.04 [M + Na] <sup>+</sup> , 400.08 [M + NH <sub>4</sub> ] <sup>+</sup> | 5.39                       | <sup>13</sup> C-DAS                        | 2     | 0.9935 | 2         | 0.9977 | 9      | 0.9857 | 9    | 0.9922 |
| 3-ADON, 15-ADON  | 361.06 [M + Na] <sup>+</sup>                                              | 5.89                       | <sup>13</sup> C-DAS                        | 20    | 0.9983 | 20        | 0.9877 | 30     | 0.9774 | 30   | 0.9864 |
| DAS              | 389.11 [M + Na] <sup>+</sup> , 384.12 [M + NH <sub>4</sub> ] <sup>+</sup> | 8.54                       | <sup>13</sup> C-DAS                        | 1     | 0.9934 | 1         | 0.9974 | 1      | 0.9811 | 1    | 0.9953 |
| HT-2             | 447.18 [M + Na] <sup>+</sup> , 442.11 [M + NH <sub>4</sub> ] <sup>+</sup> | 9.79                       | <sup>13</sup> C-HT-2                       | 2     | 0.9989 | 2         | 0.9881 | 2      | 0.9844 | 2    | 0.9799 |
| T-2              | 489.13 [M + Na] <sup>+</sup> , 484.12 [M + NH <sub>4</sub> ] <sup>+</sup> | 10.41                      | <sup>13</sup> C-HT-2                       | 1     | 0.9863 | 1         | 0.9847 | 1      | 0.9898 | 1    | 0.9891 |
| AFB <sub>1</sub> | 313.09 [M + H] <sup>+</sup>                                               | 6.99                       | <sup>13</sup> C-HT-2                       | 4     | 0.9956 | 4         | 0.9959 | 5      | 0.9974 | 5    | 0.9941 |
| AFB <sub>2</sub> | 315.11 [M + H] <sup>+</sup>                                               | 7.37                       | <sup>13</sup> C-HT-2                       | 4     | 0.9876 | 4         | 0.9986 | 5      | 0.9881 | 5    | 0.9977 |
| AFG <sub>1</sub> | 329.12 [M + H] <sup>+</sup>                                               | 7.77                       | <sup>13</sup> C-HT-2                       | 4     | 0.9934 | 4         | 0.9968 | 5      | 0.9887 | 5    | 0.9846 |
| AFG <sub>2</sub> | 331.11 [M + H] <sup>+</sup>                                               | 8.15                       | <sup>13</sup> C-HT-2                       | 4     | 0.9931 | 4         | 0.9891 | 5      | 0.9991 | 5    | 0.9908 |
| FB <sub>1</sub>  | 722.43 [M + H] <sup>+</sup>                                               | 10.03                      | <sup>13</sup> C-FB1                        | 50    | 0.9988 | 50        | 0.9965 | 65     | 0.9899 | 65   | 0.9931 |
| FB <sub>2</sub>  | 706.41 [M + H] <sup>+</sup>                                               | 10.75                      | <sup>13</sup> C-FB2                        | 25    | 0.9975 | 25        | 0.9895 | 40     | 0.9974 | 40   | 0.9844 |
| FB <sub>3</sub>  | 706.41 [M + H] <sup>+</sup>                                               | 10.47                      | <sup>13</sup> C-FB3                        | 25    | 0.9970 | 25        | 0.9947 | 40     | 0.9849 | 40   | 0.9976 |
| HFB <sub>1</sub> | 406.10 [M + H] <sup>+</sup>                                               | 9.89                       | <sup>13</sup> C-DAS                        | 6     | 0.9932 | 6         | 0.9955 | 12     | 0.9967 | 12   | 0.9913 |
| OTA              | 404.11 [M + H] <sup>+</sup> , 402.07 [M – H] <sup>–</sup>                 | 10.68                      | <sup>13</sup> C-DAS or <sup>13</sup> C-ZEN | 4     | 0.9950 | 4         | 0.9895 | 7      | 0.9996 | 7    | 0.9901 |
| OTB              | 370.09 [M + Na] <sup>+</sup>                                              | 10.12                      | <sup>13</sup> C-DAS                        | 4     | 0.9951 | 4         | 0.9963 | 7      | 0.9971 | 7    | 0.9890 |
| ZEN              | 317.10 [M – H] <sup>–</sup>                                               | 10.54                      | <sup>13</sup> C-ZEN                        | 2     | 0.9928 | 2         | 0.9947 | 2      | 0.9963 | 2    | 0.9936 |
| $\alpha$ -ZOL    | 319.12 [M – H] <sup>–</sup>                                               | 10.40                      | <sup>13</sup> C-ZEN                        | 2     | 0.9970 | 2         | 0.9991 | 2      | 0.9932 | 2    | 0.9932 |
| $\beta$ -ZOL     | 319.12 [M – H] <sup>–</sup>                                               | 9.94                       | <sup>13</sup> C-ZEN                        | 2     | 0.9917 | 2         | 0.9985 | 2      | 0.9910 | 2    | 0.9899 |
| Enn-A            | 699.31 [M + NH <sub>4</sub> ] <sup>+</sup>                                | 12.25                      | <sup>13</sup> C-DAS                        | 1     | 0.9972 | 1         | 0.9999 | 1      | 0.9941 | 1    | 0.9964 |
| Enn-A1           | 685.39 [M + NH <sub>4</sub> ] <sup>+</sup>                                | 12.37                      | <sup>13</sup> C-DAS                        | 1     | 0.9991 | 1         | 0.9989 | 1      | 0.9913 | 1    | 0.9879 |
| Enn-B            | 657.32 [M + NH <sub>4</sub> ] <sup>+</sup>                                | 12.00                      | <sup>13</sup> C-DAS                        | 1     | 0.9977 | 1         | 0.9871 | 1      | 0.9842 | 1    | 0.9997 |
| Enn-B1           | 671.31 [M + NH <sub>4</sub> ] <sup>+</sup>                                | 12.14                      | <sup>13</sup> C-DAS                        | 1     | 0.9978 | 1         | 0.9921 | 1      | 0.9960 | 1    | 0.9876 |

**Table S2.** Low, Middle and High values of recovery (R) rates & relative standard deviation (RSD) for the investigated mycotoxins in the investigated matrices.

| Matrix<br>Mycotoxin               | Wheat |      |        |      |      |      | Triticale |      |        |      |      |      | Oats |      |        |      |      |      | Barley |      |        |      |      |      |
|-----------------------------------|-------|------|--------|------|------|------|-----------|------|--------|------|------|------|------|------|--------|------|------|------|--------|------|--------|------|------|------|
|                                   | Low   | RSD  | Middle | RSD  | High | RSD  | Low       | RSD  | Middle | RSD  | High | RSD  | Low  | RSD  | Middle | RSD  | High | RSD  | Low    | RSD  | Middle | RSD  | High | RSD  |
|                                   | %     |      |        |      |      |      |           |      |        |      |      |      |      |      |        |      |      |      |        |      |        |      |      |      |
| DON <sup>a</sup>                  | 103   | 7.4  | 103    | 3.5  | 104  | 4.2  | 79        | 2.4  | 93     | 9.2  | 87   | 10.3 | 116  | 3.4  | 100    | 16.9 | 104  | 12.9 | 82     | 13.0 | 105    | 11.0 | 99   | 17.3 |
| DON-3G <sup>b</sup>               | 47    | 17.5 | 58     | 2.0  | 64   | 9.2  | 56        | 3.9  | 48     | 11.9 | 48   | 9.4  | 58   | 10.6 | 52     | 20.0 | 50   | 18.1 | 41.2   | 14.7 | 56     | 21.3 | 48   | 10.6 |
| FUS-X <sup>a</sup>                | 86    | 9.4  | 115    | 12.5 | 102  | 2.9  | 88        | 2.6  | 92     | 6.8  | 93   | 8.4  | 111  | 13.5 | 117    | 11.5 | 94   | 9.7  | 117    | 8.2  | 109    | 7.9  | 80   | 18.4 |
| NEO <sup>c</sup>                  | 102   | 10.7 | 96     | 13.3 | 93   | 3.2  | 88        | 22.0 | 92     | 5.9  | 95   | 4.1  | 109  | 8.3  | 115    | 6.1  | 95   | 10.1 | 93     | 18.5 | 110    | 6.4  | 83   | 10.3 |
| Sum of 3-and 15-ADON <sup>a</sup> | 98    | 18.6 | 115    | 7.1  | 102  | 10.6 | 83        | 15.5 | 102    | 5.7  | 93   | 12.2 | 107  | 5.7  | 89     | 16.3 | 92   | 12.1 | 89     | 19.7 | 90     | 6.0  | 103  | 15.2 |
| DAS <sup>d</sup>                  | 110   | 16.6 | 106    | 13.5 | 109  | 11.1 | 95        | 11.8 | 100    | 9.8  | 106  | 17.0 | 114  | 5.4  | 116    | 5.6  | 94   | 5.9  | 114    | 16.2 | 115    | 1.1  | 106  | 5.5  |
| HT-2 Toxin <sup>c</sup>           | 91    | 10.7 | 115    | 5.4  | 109  | 5.9  | 85        | 6.2  | 92     | 9.9  | 112  | 6.4  | 96   | 7.9  | 116    | 13.7 | 95   | 6.5  | 100    | 16.5 | 113    | 7.3  | 103  | 11.3 |
| T-2 Toxin <sup>c</sup>            | 79    | 10.2 | 111    | 8.4  | 107  | 15.4 | 93        | 9.1  | 85     | 6.1  | 121  | 5.5  | 116  | 5.8  | 104    | 17.3 | 87   | 11.8 | 109    | 4.1  | 118    | 9.4  | 108  | 14.0 |
| FB <sub>1</sub> <sup>a</sup>      | 56    | 15.3 | 46     | 17.1 | 58   | 13.0 | 59        | 15.1 | 41     | 10.2 | 53   | 6.8  | 54   | 16.3 | 52     | 4.4  | 70   | 14.0 | 74     | 13.3 | 62     | 5.1  | 58   | 18.0 |
| FB <sub>2</sub> <sup>a</sup>      | 67    | 16.6 | 78     | 5.1  | 63   | 3.9  | 91        | 15.2 | 89     | 16.3 | 89   | 9.5  | 72   | 16.4 | 67     | 17.9 | 75   | 14.6 | 72     | 9.2  | 64     | 12.1 | 77   | 6.1  |
| FB <sub>3</sub> <sup>a</sup>      | 67    | 14.4 | 82     | 10.2 | 72   | 7.6  | 60        | 14.8 | 89     | 12.8 | 80   | 8.9  | 80   | 14.4 | 75     | 18.7 | 70   | 15.8 | 66     | 10.7 | 71     | 4.3  | 80   | 10.3 |
| HFB <sub>1</sub> <sup>e</sup>     | 69    | 11.0 | 46     | 17.1 | 75   | 6.1  | 85        | 5.5  | 77     | 5.3  | 63   | 10.5 | 76   | 11.5 | 85     | 22.2 | 103  | 8.6  | 78     | 8.7  | 65     | 11.8 | 92   | 16.1 |
| OTA <sup>f</sup>                  | 110   | 3.4  | 104    | 12.4 | 100  | 12.4 | 99        | 13.0 | 92     | 13.8 | 108  | 17.1 | 100  | 7.6  | 113    | 7.7  | 119  | 6.7  | 116    | 14.0 | 111    | 3.9  | 96   | 12.5 |
| OTB <sup>f</sup>                  | 122   | 14.8 | 122    | 5.9  | 110  | 9.4  | 124       | 13.4 | 115    | 5.5  | 119  | 7.7  | 107  | 7.9  | 111    | 7.6  | 117  | 6.8  | 118    | 16.6 | 117    | 11.5 | 101  | 8.9  |
| AFB <sub>1</sub> <sup>g</sup>     | 85    | 7.6  | 93     | 9.4  | 79   | 10.3 | 73        | 8.1  | 94     | 3.8  | 72   | 10.1 | 108  | 10.8 | 92     | 7.5  | 75   | 10.6 | 71     | 13.8 | 71     | 17.4 | 96   | 8.8  |
| AFB <sub>2</sub> <sup>g</sup>     | 116   | 8.9  | 117    | 9.7  | 105  | 4.5  | 95        | 6.5  | 107    | 9.8  | 99   | 12.6 | 102  | 8.3  | 104    | 14.0 | 88   | 9.0  | 114    | 11.1 | 119    | 4.5  | 89   | 19.3 |
| AFG <sub>1</sub> <sup>g</sup>     | 113   | 9.4  | 114    | 9.2  | 93   | 7.3  | 85        | 5.3  | 93     | 10.6 | 75   | 11.2 | 84   | 22.0 | 100    | 6.7  | 86   | 15.0 | 98     | 17.0 | 104    | 16.5 | 74   | 11.2 |
| AFG <sub>2</sub> <sup>g</sup>     | 115   | 10.5 | 114    | 14.6 | 99   | 16.2 | 89        | 3.0  | 103    | 12.1 | 106  | 9.1  | 93   | 11.2 | 108    | 14.3 | 98   | 9.0  | 106    | 11.2 | 97     | 13.1 | 109  | 7.0  |
| Enn-A <sup>f</sup>                | 118   | 16.1 | 87     | 8.1  | 114  | 5.7  | 94        | 8.3  | 88     | 8.2  | 88   | 9.1  | 93   | 11.2 | 108    | 14.3 | 98   | 9.0  | 88     | 16.2 | 119    | 14.1 | 87   | 17.0 |
| Enn-A1 <sup>f</sup>               | 124   | 17.0 | 105    | 13.1 | 113  | 4.8  | 87        | 15.3 | 78     | 16.8 | 84   | 13.8 | 102  | 5.6  | 72     | 12.9 | 99   | 11.9 | 119    | 10.3 | 111    | 9.5  | 103  | 16.4 |
| Enn-B <sup>f</sup>                | 104   | 7.4  | 109    | 4.4  | 108  | 8.6  | 90        | 15.0 | 92     | 13.6 | 82   | 9.1  | 100  | 13.5 | 111    | 10.6 | 101  | 10.8 | 119    | 8.8  | 112    | 6.7  | 116  | 12.2 |
| Enn-B1 <sup>f</sup>               | 102   | 12.7 | 108    | 4.9  | 107  | 5.6  | 85        | 14.5 | 83     | 7.2  | 80   | 12.2 | 109  | 12.4 | 90     | 7.5  | 101  | 8.9  | 107    | 18.5 | 117    | 6.7  | 103  | 14.7 |
| ZEN <sup>a</sup>                  | 101   | 6.0  | 117    | 5.4  | 117  | 5.4  | 78        | 13.6 | 108    | 7.2  | 110  | 11.4 | 97   | 15.2 | 116    | 3.2  | 123  | 4.2  | 94     | 6.3  | 105    | 2.5  | 91   | 5.3  |
| α-ZOL <sup>d</sup>                | 86    | 19.2 | 94     | 14.0 | 114  | 10.7 | 100       | 24.0 | 94     | 8.7  | 103  | 10.8 | 83   | 10.9 | 112    | 4.9  | 104  | 13.9 | 65     | 10.6 | 78     | 4.3  | 106  | 9.0  |
| β-ZOL <sup>d</sup>                | 91    | 18.7 | 89     | 7.3  | 103  | 9.3  | 72        | 16.0 | 109    | 6.3  | 99   | 13.8 | 92   | 10.0 | 101    | 10.3 | 106  | 3.0  | 67     | 17.4 | 84     | 12.9 | 98   | 8.9  |

Spiking levels: <sup>a</sup> low = 200 µg·kg<sup>-1</sup>; middle = 250 µg·kg<sup>-1</sup>; high = 300 µg·kg<sup>-1</sup>; <sup>b</sup> low = 50 µg·kg<sup>-1</sup>; middle = 62.5 µg·kg<sup>-1</sup>; high = 75 µg·kg<sup>-1</sup>; <sup>c</sup> low = 100 µg·kg<sup>-1</sup>; middle = 125 µg·kg<sup>-1</sup>; high = 150 µg·kg<sup>-1</sup>; <sup>d</sup> low = 40 µg·kg<sup>-1</sup>; middle = 50 µg·kg<sup>-1</sup>; high = 60 µg·kg<sup>-1</sup>; <sup>e</sup> low = 25 µg·kg<sup>-1</sup>; middle = 31,25 µg·kg<sup>-1</sup>; high = 38 µg·kg<sup>-1</sup>; <sup>f</sup> low = 20 µg·kg<sup>-1</sup>; middle = 25 µg·kg<sup>-1</sup>; high = 30 µg·kg<sup>-1</sup>; <sup>g</sup> low = 4 µg·kg<sup>-1</sup>; middle = 5 µg·kg<sup>-1</sup>; high = 6 µg·kg<sup>-1</sup>.

**Table S3.** Results of some inter-laboratory proficiency tests (organized in 2014–2015), in which the developed method was assessed (selected mycotoxins in selected grain matrices).

| Proficiency Test                   | Compound                      | Matrix      | Assigned Value ( $\mu\text{g}\cdot\text{kg}^{-1}$ ) | Determined Value ( $\mu\text{g}\cdot\text{kg}^{-1}$ ) | z-Score Value | Result       |
|------------------------------------|-------------------------------|-------------|-----------------------------------------------------|-------------------------------------------------------|---------------|--------------|
| Romer Labs CSSMY006-M14421Z (2014) | ZEN <sup>1</sup>              | Wheat       | 119                                                 | 133                                                   | 0.5           | satisfactory |
|                                    | AFB <sub>1</sub> <sup>2</sup> |             | 5.09                                                | 5.43                                                  | 0.3           | satisfactory |
|                                    | AFs (Total) <sup>3</sup>      |             | 5.21                                                | 5.43                                                  | 0.4           | satisfactory |
| FAPAS 04260 (2015)                 | DON <sup>4</sup>              | Maize       | 1191                                                | 946                                                   | −1.3          | satisfactory |
|                                    | OTA <sup>5</sup>              |             | 3.29                                                | 4.25                                                  | 1.3           | satisfactory |
|                                    | ZEN                           |             | 200                                                 | 221                                                   | 0.5           | satisfactory |
|                                    | T-2 <sup>6</sup>              |             | 280                                                 | 330                                                   | 0.9           | satisfactory |
| FAPAS 22116 (2015)                 | HT-2 <sup>7</sup>             | Animal feed | 411                                                 | 502                                                   | 1.2           | satisfactory |
|                                    | T-2 + HT-2                    |             | 693                                                 | 832                                                   | 1.2           | satisfactory |
|                                    |                               |             |                                                     |                                                       |               |              |
| Romer Labs CSSMY008-M15421A (2015) | AFB <sub>1</sub>              | Maize       | 6.0                                                 | 5.46                                                  | −0.4          | satisfactory |
|                                    | AFs (Total)                   |             | 6.4                                                 | 6.03                                                  | −0.3          | satisfactory |

<sup>1</sup> ZEN—zearalenone, <sup>2</sup> AFB<sub>1</sub>—B<sub>1</sub> aflatoxin, <sup>3</sup> AFs (Total)—sum of aflatoxins, <sup>4</sup> DON—deoxynivalenol, <sup>5</sup> OTA—ochratoxin A, <sup>6</sup> T-2—T-2 toxin, <sup>7</sup> HT-2—HT-2 toxin