# Potential and Limitations of Grasslands $\alpha$-diversity Prediction Using Fine-Scale Hyperspectral Imagery

**Hafiz Ali Imran [1,2,*], Damiano Gianelle [1], Michele Scotton [3], Duccio Rocchini [4,5], Michele Dalponte [1], Stefano Macolino [3], Karolina Sakowska [6,7], Cristina Pornaro [3], and Loris Vescovo [1]**

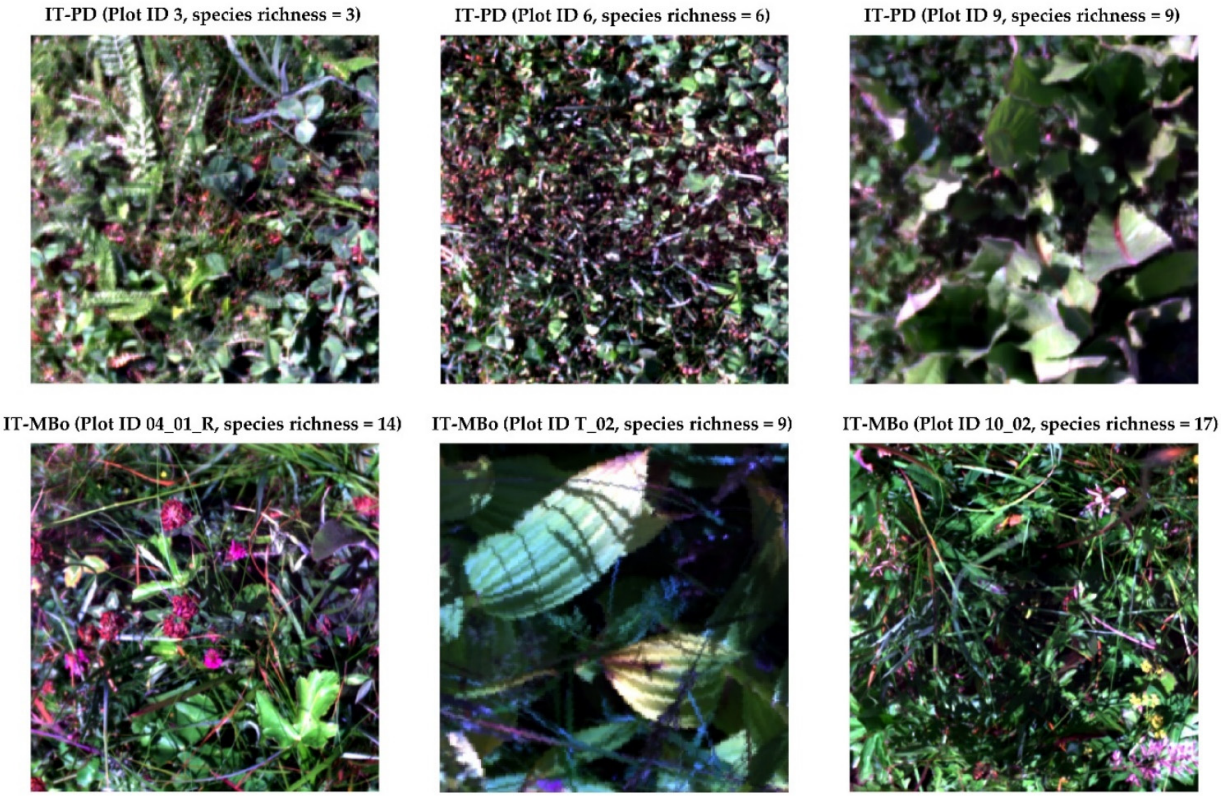


**Figure S1**. RGB images of the different plots at the IT-PD and the IT-MBo study sites.

**Table S1.** Species richness and composition of each plot investigated at the IT-PD study site.

| Plot | Species richness | Species |
|---|---|---|
| 1 | 1 | *Trifolium repens nano* |
| 2 | 2 | *Festuca rubra, Trifolium repens nano* |
| 3 | 3 | *Achillea millefolium, Festuca rubra, Trifolium repens nano* |
| 4 | 4 | *Lolium perenne, Trifolium repens nano, Cicoria selvatica, Poa pratensis* |
| 5 | 5 | *Lolium perenne, Trifolium repens nano, Cicoria selvatica, Poa pratensis, Medicago lupulina* |
| 6 | 6 | *Lolium perenne, Trifolium repens nano, Cicoria selvatica, Poa pratensis, Medicago lupulina, Poa annua* |
| 7 | 7 | *Lolium perenne, Trifolium repens nano, Cicoria selvatica, Poa pratensis, Medicago lupulina, Poa annua, Festuca arundinacea* |
| 8 | 8 | *Lolium perenne, Trifolium repens nano, Cicoria selvatica, Poa pratensis, Medicago lupulina, Poa annua, Festuca arundinacea, Taraxacum officinale* |
| 9 | 9 | *Lolium perenne, Trifolium repens nano, Cicoria selvatica, Poa pratensis, Medicago lupulina, Poa annua, Festuca arundinacea, Taraxacum officinale, Lotus corniculatus* |

**Table S2.** Species richness and composition of each of the 25 plots investigated at the IT-MBo study site.

| Plot | Species richness | Species |
|---|---|---|
| 01_2 | 16 | *Agrostis tenuis, Brachypodium pinnatum (rup), Briza media Chamaecytisus hirsutus, Euphrasia rostkoviana, Festuca nigrescens, Galium pumilum, Lotus corniculatus, Nardus stricta, Phleum alpinum, Polygonum viviparum, Potentilla crantzii, Potentilla erecta, Ranunculus montanus, Trifolium pratense, Trifolium repens* |
| 02_2 | 13 | *Achillea millefolium, Agrostis tenuis, Crocus albiflorus, Euphrasia rostkoviana, Galium pumilum, Hieracium cymosum, Nardus stricta, Pimpinella major, Polygonum viviparum, Potentilla erecta, Ranunculus montanus, Trifolium pratense, Trifolium repens* |
| 04_1_R | 14 | *Achillea millefolium, Anthoxanthum odoratum, Brachypodium pinnatum (rup), Chaerophyllum hirsutum, Galium pumilum, Helianthemum numm. grandifl., Laserpitium krapfii ssp. Gaudinii, Lathyrus pratensis, Nardus stricta, Paradisea liliastrum, Phyteuma betonicifolium, Polygonum viviparum, Potentilla erecta, Trifolium pratense* |
| 05_2 | 12 | *Achillea millefolium, Agrostis tenuis, Festuca nigrescens, Hypericum maculatum, Lathyrus pratensis, Lotus corniculatus, Nardus stricta, Plantago atrata, Polygonum viviparum, Potentilla erecta, Ranunculus montanus, Trifolium repens* |
| 06_2 | 13 | *Alchemilla vulgaris, Festuca nigrescens, Gentianella germanica, Leucanthemum vulgare, Lotus corniculatus, Nardus stricta, Plantago atrata, Poa violacea, Polygonum viviparum, Potentilla erecta, Rhinanthus alectorolophus, Trifolium montanum, Trifolium repens* |
| 07_2 | 12 | *Carex montana, Cerastium caespitosum, Festuca nigrescens, Helianthemum numm. grandifl., Lathyrus pratensis, Phyteuma orbiculare, Plantago atrata, Pulsatilla alpina ssp. Alpina, Rumex alpestris, Stachys alopecuros, Trifolium pratense, Vicia cracca* |
| 08_2 | 9 | *Agrostis tenuis, Chamaecytisus hirsutus, Festuca nigrescens, Genista germanica, Luzula campestris, Nardus stricta, Plantago atrata, Polygonum viviparum, Potentilla erecta* |
| 09_2 | 12 | *Achillea millefolium, Agrostis tenuis, Chamaecytisus hirsutus, Festuca nigrescens, Helianthemum numm. grandifl., Hieracium cymosum, Lotus corniculatus, Plantago atrata, Polygonum viviparum, Ranunculus montanus, Stachys alopecuros, Trifolium montanum* |

| | | |
|---|---|---|
| 10_2 | 17 | *Agrostis tenuis, Alchemilla vulgaris, Crocus albiflorus, Festuca nigrescens, Galium pumilum, Hieracium pilosella, Laserpitium krapfii ssp. Gaudinii, Lathyrus pratensis, Leontodon hispidus, Polygonum viviparum, Potentilla erecta, Ranunculus montanus, Scorzonera aristata, Trifolium montanum, Trifolium pratense, Trifolium repens, Trollius europaeus* |
| 11_2 | 9 | *Achillea millefolium, Brachypodium pinnatum (rup), Briza media, Genista tinctoria, Lilium martagon, Phyteuma betonicifolium, Pulsatilla alpina ssp. Alpina, Ranunculus montanus, Trifolium pratense* |
| 12_2 | 15 | *Agrostis tenuis, Cerastium caespitosum, Chamaecytisus hirsutus, Crocus albiflorus, Festuca nigrescens, Galium pumilum, Galium rubrum, Genista tinctoria, Geum montanum, Nardus stricta, Paradisea liliastrum, Polygonum viviparum, Pulsatilla alpina ssp. Alpina, Vaccinium myrtillus, Viola canina* |
| 13_2 | 16 | *Arnica montana, Chaerophyllum hirsutum, Crocus albiflorus, Euphrasia rostkoviana, Galium pumilum, Geum montanum, Nardus stricta, Phleum alpinum, Phyteuma betonicifolium, Plantago atrata, Polygonum viviparum, Potentilla erecta, Ranunculus montanus, Stellaria graminea, Trifolium pratense, Trifolium repens* |
| 14_2 | 14 | *Achillea millefolium, Agrostis tenuis, Cerastium caespitosum, Chaerophyllum hirsutum, Dactylis glomerata, Festuca nigrescens, Hypericum maculatum, Phleum alpinum, Poa violacea, Polygonum viviparum, Ranunculus montanus, Rumex alpestris, Stellaria graminea, Trifolium repens* |
| 16_2 | 15 | *Agrostis tenuis, Chaerophyllum hirsutum, Chamaecytisus hirsutus, Galium pumilum, Nardus stricta, Paradisea liliastrum, Phleum alpinum, Phyteuma betonicifolium, Polygonum viviparum, Potentilla erecta, Pulsatilla alpina ssp. Alpina, Ranunculus montanus, Stellaria graminea, Trifolium repens, Trollius europaeus* |
| 17_2 | 15 | *Agrostis tenuis, Alchemilla vulgaris, Campanula scheuchzeri, Chaerophyllum hirsutum, Dactylis glomerata, Festuca nigrescens, Nardus stricta, Poa pratensis, Polygonum viviparum, Potentilla crantzii, Ranunculus montanus, Trifolium pratense, Trifolium repens, Valeriana wallrothii, Veronica chamaedrys* |
| 18_2 | 11 | *Achillea millefolium, Agrostis tenuis, Brachypodium pinnatum (rup), Briza media, Chamaecytisus hirsutus, Crocus albiflorus, Dactylis glomerata, Hypericum maculatum, Phleum alpinum, Polygonum viviparum, Trifolium repens* |

| | | |
|---|---|---|
| 19_2 | 15 | *Agrostis tenuis, Campanula scheuchzeri, Cerastium caespitosum, Chaerophyllum hirsutum, Crocus albiflorus, Euphrasia rostkoviana, Festuca nigrescens, Geum montanum, Plantago atrata, Polygonum viviparum, Potentilla crantzii, Potentilla erecta, Ranunculus montanus, Stellaria graminea, Trifolium repens* |
| 20_2 | 16 | *Agrostis tenuis, Brachypodium pinnatum (rup), Centaurea triumfettii, Festuca nigrescens, Helianthemum numm. grandifl., Heracleum sphondylium L., Koeleria pyramidata, Phyteuma orbiculare, Plantago atrata, Poa violacea, Polygonum viviparum, Potentilla erecta, Ranunculus montanus, Trifolium montanum, Trifolium repens, Viola canina* |
| T_02 | 9 | *Achillea millefolium, Alchemilla vulgaris, Brachypodium pinnatum (rup), Briza media, Chamaecytisus hirsutus, Festuca nigrescens, Koeleria pyramidata, Poa chaixii, Veratrum album* |
| T_03 | 4 | *Agrostis tenuis, Carex sp., Epilobium angustifolium, Fragaria sp.* |
| T_05 | 11 | *Agrostis tenuis, Chamaecytisus hirsutus, Crocus albiflorus, Festuca nigrescens, Galium pumilum, Helianthemum numm. grandifl., Nardus stricta, Polygonum viviparum, Trifolium montanum, Trifolium pratense, Trifolium repens* |
| T_10_B | 2 | *Agrostis tenuis, Chaerophyllum hirsutum* |
| T_14 | 16 | *Achillea millefolium, Agrostis tenuis, Festuca nigrescens, Festuca pratensis, Galium spp., Leontodon hispidus, Leucanthemum vulgare, Phyteuma betonicifolium, Plantago atrata, Ranunculus montanus, Rhinanthus alectorolophus, Stellaria graminea, Trifolium pratense, Trifolium repens, Trisetum flavescens, Veronica chamaedrys* |
| T_15 | 14 | *Achillea millefolium, Agrostis tenuis, Dactylis glomerata, Festuca nigrescens, Festuca pratensis, Galium spp., Gentiana kochiana, Gentiana lutea, Leontodon hispidus, Plantago atrata, Trifolium pratense, Trifolium repens, Trisetum flavescens, Veronica chamaedrys* |
| T_20 | 10 | *Agropyron repens, Alchemilla vulgaris, Alopecurus pratensis, Dactylis glomerata, Festuca pratensis, Ranunculus acris, Rumex obtusifolius, Taraxacum officinale, Trisetum flavescens, Vicia sepium* |

**Table S3.** The highest Pearson correlation coefficient (R) and *p*-values (in brackets) for the relationship between species richness and two optical diversity metrics (CV and SD) for different processing levels at the IT-PD study site. The highest R values for each processing level are highlighted in bold.

| **Processing levels** | **IT-PD 1 mm** | | | | **IT-PD 1.5 mm** | | | |
|---|---|---|---|---|---|---|---|---|
| | **WL**[1] | **CV**[1] | **WL** | **SD**[1] | **WL** | **CV** | **WL** | **SD** |
| Level$_0$ | 560 | 0.60 (0.086) | 703 | 0.42 (0.265) | 435 | 0.71 (0.031) | 685 | **0.74 (0.023)** |
| Level$_1$ | 563 | 0.58 (0.102) | 733 | 0.49 (0.179) | 432 | 0.78 (0.014) | 927 | **0.78 (0.013)** |
| Level$_2$ | 927 | 0.83 (0.006) | 927 | **0.84 (0.005)** | 435 | 0.84 (0.005) | 927 | 0.80 (0.011) |
| Level$_3$ | 452 | 0.67 (0.048) | 455 | 0.67 (0.049) | 412 | **0.87 (0.003)** | 412 | 0.86 (0.003) |

[1] WL: wavelength, CV: coefficient of variation, SD: standard deviation.

**Table S4.** The highest Pearson correlation coefficient (R) and *p*-values (in brackets) for the relationship between biodiversity indices (species richness, Shannon's index, species evenness, and Simpson's index) and the two optical diversity metrics (CV and SD) for different processing levels at the IT-MBo study site. The highest R values for each processing level are highlighted in bold.

| **Processing levels** | **Richness** | | | | **Shannon** | | | | **Evenness** | | | | **Simpson** | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | **WL**[1] | **CV**[1] | **WL** | **SD**[1] | **WL** | **CV** | **WL** | **SD** | **WL** | **CV** | **WL** | **SD** | **WL** | **CV** | **WL** | **SD** |
| $Level_0$ | 420 | 0.25 (0.236) | 499 | **0.25 (0.224)** | 429 | -0.06 (0.772) | 499 | 0.06 (0.79) | 409 | -0.11 (0.594) | 499 | 0.03 (0.898) | 446 | -0.12 (0.576) | 499 | -0.04 (0.847) |
| $Level_1$ | 423 | 0.29 (0.154) | 429 | 0.34 (0.096) | 415 | 0.24 (0.24) | 418 | 0.32 (0.118) | 415 | 0.24 (0.24) | 418 | **0.35 (0.091)** | 409 | 0.16 (0.459) | 409 | 0.21 (0.308) |
| $Level_2$ | 409 | 0.25 (0.236) | 682 | 0.43 (0.03) | 409 | 0.24 (0.248) | 685 | **0.48 (0.018)** | 409 | 0.22 (0.294) | 685 | 0.47 (0.019) | 409 | 0.20 (0.344) | 409 | 0.35 (0.084) |
| $Level_3$ | 930 | 0.04 (0.852) | 679 | 0.5 (0.011) | 911 | 0.10 (0.652) | 688 | 0.55 (0.004) | 911 | 0.05 (0.816) | 688 | **0.56 (0.004)** | 920 | 0.15 (0.479) | 691 | 0.4 (0.047) |

[1] WL: wavelength, CV: coefficient of variation, SD: standard deviation.

**Table S5.** The highest Pearson correlation coefficient (R) and *p*-values (in brackets) for the relationship between species richness and the two optical diversity metrics (CV and SD) calculated from Level3 processed data at different spatial scales for the IT-PD study site. The highest R values for each spatial scale are highlighted in bold.

| Spatial scale | IT-PD 1 mm | | | |
|---|---|---|---|---|
| | WL[1] | CV[1] | WL | SD[1] |
| 1 mm | 452 | **0.67 (0.05)** | 455 | **0.67 (0.05)** |
| 2.5 mm | 452 | **0.67 (0.05)** | 455 | **0.67 (0.05)** |
| 5 mm | 554 | **0.68 (0.05)** | 455 | 0.66 (0.054) |
| 1 cm | 452 | 0.73 (0.027) | 452 | **0.74 (0.023)** |
| 2.5 cm | 412 | 0.65 (0.059) | 688 | **0.79 (0.011)** |
| 5 cm | 412 | 0.42 (0.257) | 694 | **0.73 (0.027)** |
| 8.3 cm | 412 | 0.11 (0.769) | 412 | **0.12 (0.762)** |

[1] WL: wavelength, CV: coefficient of variation, SD: standard deviation.

**Table S6**. The highest Pearson correlation coefficient (R) and *p*-values (in brackets) for the relationship between biodiversity indices (species richness, Shannon's index, species evenness, and Simpson's index) and two optical diversity metrics (CV and SD) calculated from Level3 processed data at different spatial scales for the IT-MBo study site. The highest R values for each spatial scale are highlighted in bold.

| **Spatial** | **Richness** | | | | **Shannon** | | | | **Evenness** | | | | **Simpson** | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **scale** | **WL[1]** | **CV[1]** | **WL** | **SD[1]** | **WL** | **CV** | **WL** | **SD** | **WL** | **CV** | **WL** | **SD** | **WL** | **CV** | **WL** | **SD** |
| 1 mm | 930 | 0.05 (0.833) | 679 | 0.50 (0.012) | 914 | 0.10 (0.643) | 685 | **0.55 (0.005)** | 911 | 0.05 (0.807) | 688 | 0.55 (0.004) | 921 | 0.15 (0.482) | 691 | 0.39 (0.054) |
| 2.5 mm | 930 | 0.03 (0.875) | 679 | 0.48 (0.016) | 921 | 0.08 (0.715) | 688 | **0.54 (0.005)** | 914 | 0.03 (0.881) | 688 | **0.54 (0.005)** | 921 | 0.13 (0.549) | 688 | 0.38 (0.062) |
| 5 mm | 930 | 0.05 (0.828) | 682 | 0.42 (0.035) | 914 | 0.07 (0.737) | 685 | **0.48 (0.016)** | 911 | 0.02 (0.913) | 685 | **0.48 (0.016)** | 930 | 0.10 (0.628) | 688 | 0.33 (0.105) |
| 1 cm | 412 | 0.00 (0.996) | 682 | 0.33 (0.108) | 908 | 0.03 (0.877) | 682 | **0.35 (0.085)** | 908 | -0.01 (0.955) | 685 | 0.35 (0.088) | 908 | 0.03 (0.894) | 688 | 0.23 (0.274) |
| 2.5 cm | 412 | 0.09 (0.653) | 685 | **0.38 (0.063)** | 917 | 0.02 (0.934) | 685 | 0.33 (0.11) | 917 | 0.02 (0.919) | 688 | 0.29 (0.154) | 685 | -0.04 (0.84) | 688 | 0.22 (0.3) |
| 5 cm | 682 | -0.02 (0.942) | 685 | **0.15 (0.466)** | 911 | -0.07 (0.729) | 685 | 0.03 (0.907) | 911 | -0.04 (0.852) | 685 | 0.01 (0.953) | 914 | -0.19 (0.37) | 552 | -0.13 (0.523) |
| 8.3 cm | 911 | **0.25 (0.23)** | 911 | **0.25 (0.23)** | 911 | 0.19 (0.352) | 911 | 0. 2 (0.338) | 911 | 0.18 (0.396) | 911 | 0.19 (0.375) | 426 | 0.11 (0.604) | 426 | 0.10 (0.644) |

[1] WL: wavelength, CV: coefficient of variation, SD: standard deviation.

**Table S7.** The highest Pearson correlation coefficient (R) and *p*-values (in brackets) for the relationship between species richness and the two optical diversity metrics (CV and SD) calculated from Level$_3$ processed data at different sample size for the IT-PD study site. The highest R values are highlighted in bold.

| Sub-sample (No. of pixels) | IT-PD 1 mm | | | |
|---|---|---|---|---|
| | WL[1] | CV[1] | WL | SD[1] |
| All pixels | 452 | 0.67 (0.042) | 455 | **0.67 (0.048)** |
| 500 pixels | 554 | **0.71 (0.034)** | 452 | 0.66 (0.054) |
| 300 pixels | 452 | 0.68 (0.046) | 452 | **0.70 (0.036)** |
| 250 pixels | 473 | **0.63 (0.071)** | 490 | 0.61 (0.079) |
| 200 pixels | 452 | 0.64 (0.065) | 452 | **0.67 (0.048)** |
| 150 pixels | 679 | 0.62 (0.078) | 691 | **0.71 (0.033)** |
| 100 pixels | 546 | **0.69 (0.042)** | 706 | 0.66 (0.054) |
| 50 pixels | 412 | **0.77 (0.015)** | 412 | 0.77 (0.016) |

[1] WL: wavelength, CV: coefficient of variation, SD: standard deviation.

**Table S8.** The highest Pearson correlation coefficient (R) and *p*-values (in brackets) for the relationship between biodiversity indices (species richness, Shannon's index, species evenness, and Simpson's) and the two optical diversity metrics (CV and SD) calculated from Level3 processed data at different sample size for the IT-MBo study site. The highest R values are highlighted in bold.

| **Sub-sample** | **Richness** | | | | **Shannon** | | | | **Evenness** | | | | **Simpson** | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **(No. of pixels)** | **WL[1]** | **CV[1]** | **WL** | **SD[1]** | **WL** | **CV** | **WL** | **SD** | **WL** | **CV** | **WL** | **SD** | **WL** | **CV** | **WL** | **SD** |
| All pixels | | 0.04 | | 0.50 | | 0.10 | | 0.55 | | 0.05 | | **0.56** | | 0.15 | | 0.40 |
| | 930 | (0.852) | 679 | (0.011) | 911 | (0.652) | 688 | (0.004) | 911 | (0.816) | 688 | **(0.004)** | 920 | (0.479) | 691 | (0.047) |
| 500 pixels | | 0.03 | | 0.35 | | 0.13 | | 0.42 | | 0.09 | | 0.43 | | 0.07 | | **0.44** |
| | 927 | (0.883) | 679 | (0.086) | 920 | (0.549) | 685 | (0.035) | 911 | (0.671) | 688 | (0.032) | 911 | (0.736) | 688 | **(0.029)** |
| 300 pixels | | 0.06 | | 0.37 | | 0.10 | | 0.45 | | 0.05 | | 0.46 | | 0.06 | | **0.47** |
| | 920 | (0.776) | 679 | (0.068) | 908 | (0.624) | 682 | (0.025) | 908 | (0.803) | 682 | (0.022) | 908 | (0.768) | 688 | **(0.018)** |
| 250 pixels | | 0.11 | | 0.50 | | 0.03 | | 0.56 | | 0.04 | | 0.59 | | 0.09 | | **0.62** |
| | 412 | (0.614) | 679 | (0.012) | 679 | (0.899) | 685 | (0.003) | 679 | (0.861) | 685 | (0.002) | 679 | (0.684) | 685 | **(0.001)** |
| 200 pixels | | 0.16 | | 0.48 | | -0.01 | | **0.55** | | -0.03 | | 0.53 | | 0.03 | | 0.52 |
| | 426 | (0.441) | 682 | (0.015) | 420 | (0.948) | 685 | **(0.005)** | 415 | (0.905) | 685 | (0.006) | 415 | (0.892) | 685 | (0.007) |
| 150 pixels | | 0.14 | | 0.45 | | 0.17 | | **0.54** | | 0.13 | | 0.53 | | 0.12 | | 0.53 |
| | 418 | (0.507) | 682 | (0.024) | 914 | (0.429) | 682 | **(0.006)** | 911 | (0.552) | 682 | (0.006) | 914 | (0.57) | 685 | (0.006) |
| 100 pixels | | 0.03 | | 0.36 | | 0.09 | | 0.41 | | 0.07 | | 0.43 | | 0.05 | | **0.43** |
| | 930 | (0.881) | 676 | (0.082) | 911 | (0.687) | 679 | (0.04) | 911 | (0.726) | 679 | (0.034) | 911 | (0.829) | 679 | **(0.034)** |
| 50 pixels | | 0.15 | | 0.32 | | 0.15 | | 0.40 | | 0.14 | | **0.42** | | 0.11 | | 0.40 |
| | 429 | (0.461) | 688 | (0.123) | 930 | (0.487) | 691 | (0.048) | 930 | (0.52) | 691 | **(0.039)** | 412 | (0.593) | 688 | (0.045) |

[1] WL: wavelength, CV: coefficient of variation, SD: standard deviation