

Article

Forecasting Young Apple Tree Bud Status with a Visible/Near-Infrared Spectrometer

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Abstract: Being able to ascertain the physiological condition of the buds on a young apple tree before bud burst could help farmers manage their orchards more efficiently, especially if they could do so without destroying the buds in the process. The experiments carried out in this study were conducted with the aim of distinguishing shoot from non-shoot buds before bud burst using a visible/near-infrared spectrometer, a device that does not destroy the buds being tested. Tests on spring-planted (April 30, 2021) trees were conducted to check shoot and non-shoot bud physiology and the winter dormancy of young ‘Jonagold’, ‘Miyabi Fuji’ and ‘Orin’ apple trees. The light absorbance of the shoot buds before bud burst was much lower than the light absorbance of the non-shoot buds as checked on the visible/near-infrared spectrometer. The highest first factor effect was determined by a PCA test conducted on shoot and non-shoot ‘Jonagold’ buds (99.9%) at a range of 640-652 nm, ‘Miyabi Fuji’ buds (99.7%) at 654-680 nm and ‘Orin’ buds (99.6%) at 704-766 nm seven days before bud burst. We also found that the highest level of accuracy, using the Classifier analysis, between shoot and non-shoot ‘Jonagold’ buds (76.6%) was one day before bud burst, for ‘Miyabi Fuji’ buds (82.1%) it was three days before and for ‘Orin’ buds (76.3%) it was two days before. These findings suggest that growers can more effectively manage the development of the young trees in their orchards with a visible/near-infrared spectrometer.

Keywords: bud burst; Classification type; shoot bud; non-shoot bud; ‘Jonagold’; ‘Miyabi Fuji’; ‘Orin’.

1. Introduction

In young apple orchards, the determining factors in their future productivity are the healthy growth of young apple trees, the number of fruit branches on each tree, and the accurate construction of the tree in order to make it possible to establish where the fruit will appear on the tree in the future. It is also essential to know the exact physiology of the buds before effective orchard management activities can be carried out. Knowing the bud's physiology and correctly evaluating it could help the grower identify whether or not a shoot will be formed from a particular pre-bud burst stage of development. This would enable him or her to carry out fieldwork in a timely way. It is not easy to distinguish the shoot buds that would continue to develop into shoots from the non-shoot buds that remain dormant. In addition, physiological changes in the buds also depend on the apple variety and climatic conditions. In this study, the bud light absorbance of ‘Jonagold’, ‘Miyabi Fuji’ and ‘Orin’ varieties were tested and analyzed.

'Jonagold' is a 'Golden Delicious' and 'Jonathan' cross that was carried out in 1943 at the New York State Agricultural Experiment Field breeding program in Geneva, New York. It was released in 1968 [5].

'Miyabi Fuji' is a bud sport of 'Fuji' having good fruit coloration [2]. The 'Fuji' cultivar is a 'Delicious' × 'Ralls Janet' cross that was bred in 1939 and has become one of the world's best-known apple varieties [3, 5, 8, 11]. It stores well (7-11 months), has good shelf life and holds its taste and surface quality [6]. In 1962, the name 'Fuji' was given to the cultivar by researchers at the Agricultural Research Station in Morioka, Japan. The name comes from the town of Fujisaki (Aomori Prefecture, Japan), where the cross was originally made.

'Orin' is a hybrid of the 'Golden Delicious' and 'Indo' cultivars. It has a yellow-green appearance, a pleasant taste, and a unique aroma." [14].

Kramer and Kozlowski [10] found that, "A bud is an embryonic axis with its appendages. Height growth results from the activity of apical meristems or growing point." One of the most practical ways to examine bud physiology is to utilize modern technologies that do so in a non-destructive way. One of these technologies is near-infrared spectroscopy that exposes material to near-infrared light (700-3000 nm). UV-visible spectroscopy is widely used to determine organic substances by exposing the objects to visible and ultraviolet light (200-800 nm) and measuring their absorbance [4, 7, 13].

Spectroscopic technologies now make it possible to identify fruit tree bud characteristics before pruning without destroying the bud. However, no research has been reported on detecting shoot and non-shoot buds using non-destructive measurement methods. A visible/near-infrared spectrometer is one such non-invasive technological tool which exposes an object to light in visible and near-infrared spectroscopy bandwidths (640-1050 nm). This spectrometer is relatively easy to use in the field and is also very efficient timewise. Therefore, in the following study, we used a visible/near-infrared spectrometer to distinguish shoot from non-shoot buds.

2. Materials and Methods

2.1 Plant materials

Shoot and non-shoot buds from three young 'Miyabi Fuji', 'Jonagold' and 'Orin' apple trees, grafted onto semi-vigorous 'Marubakaido' (*Malus prunifolia* 'Ringo') rootstocks, were used in this study. The three trees, one of each cultivar, were planted on April 30, 2021. Before planting, all saplings were scaled to the same size by cutting them to a length of 70 cm; roots were cut back to 10 cm. The young apple trees were then placed in 11 L black plastic nursery pots. All trees were purchased from "HARADA NURSERY" Co, Ltd. The experiments were conducted at the Faculty of Agriculture and Life Science on the campus of Hirosaki University. The 'Jonagold' and "Orin' buds were tested on April 30 through to May 6; the buds on the 'Miyabi Fuji' were tested on April 30 through to May 8. The 'Jonagold' and 'Orin' bud bursts were observed on May 6, 2021, whereas for 'Miyabi Fuji' it was May 8, 2021. The buds were tested successfully with an ultra-mini visible/near-infrared spectrometer in orchard conditions on each of the testing dates (Figure 1).

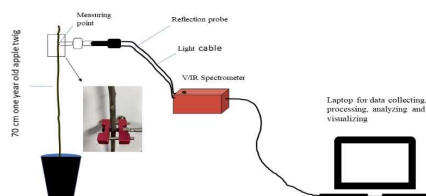


Figure-1. A close-up view of the object with an extension of the visible/near-infrared spectrometer connected to a laptop with a Spectro data fetching procedure.

2.2 Non-destructive measurement

The visible/near-infrared spectrometer is a device that measures the amount of light that passes through an object without destroying it, where some wavelengths are passed and others are absorbed. The OMT-NIR-M1 spectrometer used in this study was manufactured by Opticom Co., Ltd. using Spectral Ratio Version 1.1.0.1 software. This spectrometer measures from a range of 640 nm to 1050 nm, with an interval of 2 nm. Measurement parameters were adjusted to amp gain-high, to memory integration-16 and to smoothing points-16 nm. All buds were measured with this spectrometer and the spectral data were collected. The spectral data were then used to identify and distinguish pre-bud burst shoot buds from non-shoot buds. Twelve days after bud burst, we observed that the top four or five buds had become shoots and the buds below had become leaves or remained dormant.

2.3 Statistical analysis

The difference between the shoot and non-shoot buds of the ‘Jonagold’, ‘Miyabi Fuji’ and ‘Orin’ trees were analyzed with the student’s t-Test, and the buds Spectro data were analyzed using the R studio version 1.3.1073 (© 2009-2020 RStudio, PBC). Changes were identified on a line graph that showed the buds’ 1st and 2nd Differential at ranges of 640-652 nm, 654-680 nm, 682-702 nm, 704-766 nm, 768-924 nm and 926-1050 nm. The observations of light absorbance collected from the buds on the ‘Jonagold’, ‘Miyabi Fuji’ and ‘Orin’ trees with the visible/near-infrared spectrometer seven days before bud burst were each analyzed by principal component analysis (PCA).

Light absorbance differences for the ‘Jonagold’, ‘Miyabi Fuji’ and ‘Orin’ tree buds observed on the visible/near-infrared spectrometer were analyzed using the MATLAB R2018b version 9.5.0.1298439 Classification model (© 1984-2018 the MathWorks, Inc).

Classification was achieved with the following three steps: First of all, the Spectro data of the buds were arranged for all three varieties and for all seven test dates. After that, each new session was set as a cross-validation (folds: 5 without PCA), then the scaled data were imported into the MATLAB workspace. Finally, two wavelengths, 666 nm and 692 nm for ‘Jonagold’, 666 nm and 694 nm for ‘Miyabi Fuji’ and ‘Orin’, were chosen and the 22 algorithms were applied to classify the Spectro data for all the buds, which were then trained (Figure 2).

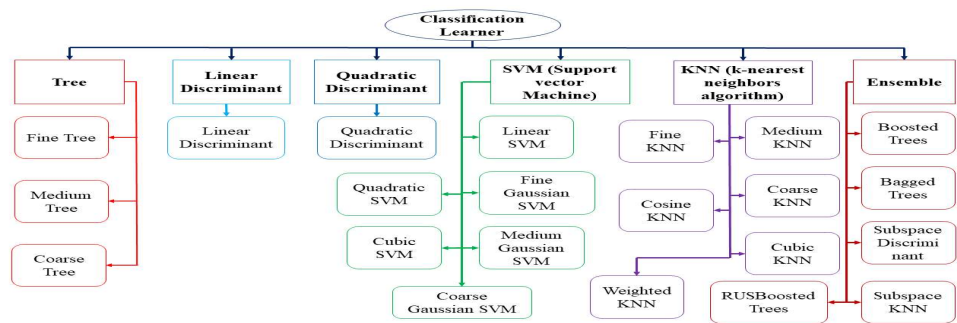


Figure-2. Classification Learner all 22 machine-learning algorithms name and related Classifier.

3. Results

3.1. Shoot and non-shoot buds

The bar chart provides information regarding the changes in the shoot and non-shoot buds of the young 'Jonagold', 'Miyabi Fuji' and 'Orin' trees after bud burst in 2021 (Figure 3). The percentage of shoot buds was lower, while the percentage of non-shoot buds was higher for all three varieties. There were about 7 'Jonagold' shoot buds, which were approximately 5 fewer than the number of non-shoot buds and significant differences in the percentages ($P \leq 0.01$) were observed. The 'Miyabi Fuji' shoot buds numbered about 6, whereas there were around 12 non-shoot buds; significant differences in the percentages ($P \leq 0.01$) were observed. However, the number of shoot buds on the 'Orin' were about 7, with the number of non-shoot buds reaching around 11; there were significant differences in the percentages ($P \leq 0.001$) between the shoot and non-shoot buds.

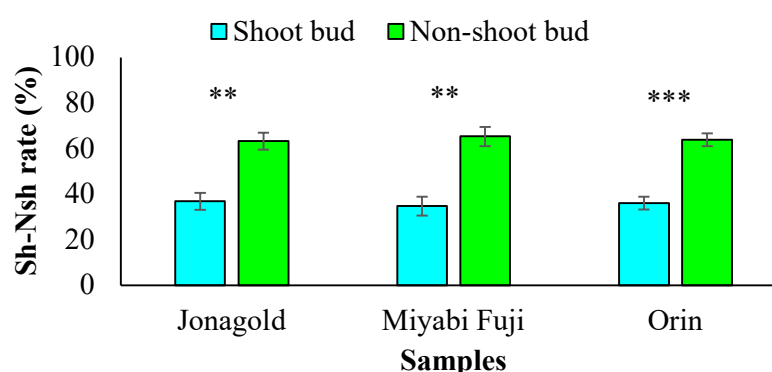


Figure-3. The changes in the shoot (Sh) and non-shoot (Nsh) buds of the young 'Jonagold', 'Miyabi Fuji' and 'Orin' trees on May 20, 2021. The percentages of the shoot and non-shoot buds were established. Means \pm standard error and significant differences between the shoot and non-shoot buds according to a T-test; (**) - Significant at 0.01, (***) - Significant at 0.001, (n=4).

3.2. Absorbances, 1st Differentials and 2nd Differentials

The line graphs in Figure 4 illustrate the light absorbance rates of the shoot and non-shoot buds. The 1st Differential and the 2nd Differential for the 'Jonagold' were observed seven days before bud burst and were measured with the visible/near-infrared spectrometer. The light absorbance of the shoot and non-shoot buds showed a downward trend (Figure 4 (a)), while the 1st and 2nd Differentials were unstable (Figure 4 (b) and (c)). During the seven days before bud burst the light absorbance rates of the shoot and non-shoot buds were nearly 1 to 0.9 ($\log(1/R)$) within the 640-672 nm range, then sharply decreased to 0.2 ($\log(1/R)$) at 1050 nm. The 1st Differential of the shoot buds was lower than the non-shoot buds at 692 nm. The 2nd Differential of the shoot buds was lower than that of the non-shoot buds at 676 nm, while it was higher at 700 nm.

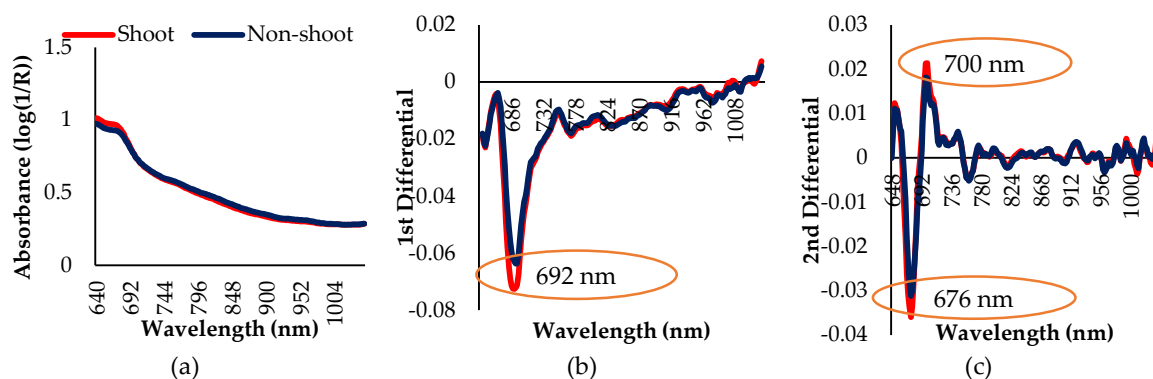


Figure-4. The light absorbance of the shoot and non-shoot buds for the 'Jonagold' seven days before bud burst. The 1st Differential and 2nd Differential on the visible/near-infrared spectrometer; (a) Absorbance ($\log(1/R)$); (b) 1st Differentials and (c) 2nd Differentials.

The line graphs in Figure 5 illustrate the the light absorbance rates of the shoot and non-shoot buds. The 1st Differential and the 2nd Differential for the 'Miyabi Fuji', seven days before bud burst, were observed with the visible/near-infrared spectrometer. The light absorbance of the shoot and non-shoot buds showed a sharply downward trend, while the light absorbance of the shoot buds was lower than that of the non-shoot buds (Figure 5 (a)). On the other hand, the 1st and 2nd Differentials were unstable (Figure 5 (b) and (c)). During the seven days before bud burst, the light absorbance rate of the shoot buds was nearly 1.01 to 0.9 ($\log(1/R)$) between 640-684 nm, and that of the non-shoot buds was 1.1-1.02 between 640-684 nm, then sharply decreased to 0.3 ($\log(1/R)$) for the shoot buds and 0.5 ($\log(1/R)$) for the non-shoot buds at 1050 nm. The 1st Differential of the shoot buds was lower than the non-shoot buds at 692 nm. The 2nd Differential of the shoot buds was lower than that of the non-shoot buds at 678 nm, while it the 2nd Differential of the shoot buds was higher than that of the non-shoot buds at 702 nm.

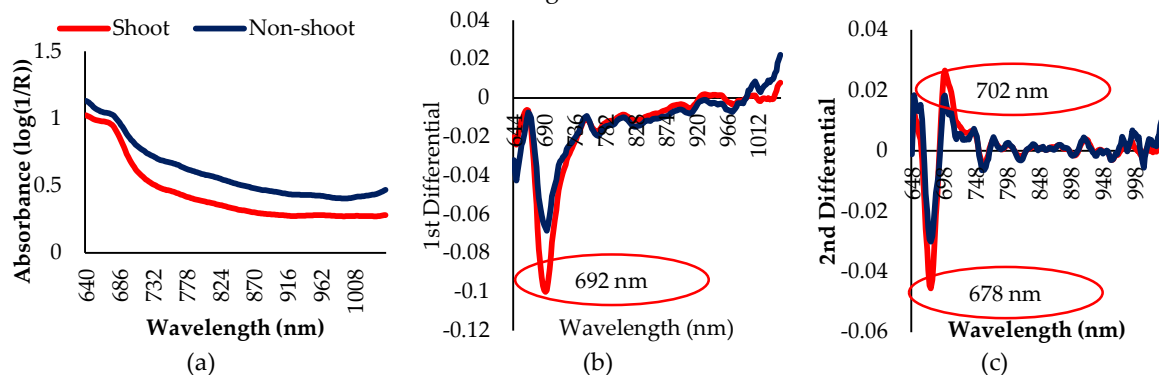


Figure-5. The light absorbance of the shoot and non-shoot buds for the 'Miyabi Fuji' seven days before bud burst. The 1st Differential and 2nd Differential on the visible/near-infrared spectrometer; (a) Absorbance ($\log(1/R)$); (b) 1st Differentials and (c) 2nd Differential.

The line graphs in Figure 6 show the light absorbance of the shoot and non-shoot buds. The, 1st Differential and 2nd Differential for the 'Orin', seven days before bud burst, were measured with the visible/near-infrared spectrometer. The light absorbance of the shoot and non-shoot buds showed a sharply descending trend, while the light absorbance of the shoot buds was lower than that of the non-shoot buds (Figure 6 (a)). On the other hand, the 1st and 2nd Differentials were unstable (Figure 6 (b) and (b)). During the seven days before bud burst, the light absorbance rate of the shoot buds was nearly 1.4 to 1.3 ($\log(1/R)$) between 640-678 nm, and that of the non-shoot buds was 1.5-1.4 between 640-678 nm, then sharply decreased to 0.4 ($\log(1/R)$) for the light absorbance of the shoot buds

and non-shoot buds at 1050 nm. The 1st Differential of the shoot buds was higher than the non-shoot buds at 688 nm. The 2nd Differential of the shoot buds was higher than that of the non-shoot buds at 676 nm, while the 2nd Differentials of the shoot and non-shoot buds were approximately equal at 700 nm.

3.3. Principal component analysis

Table 1 shows the PCA of the light absorbance of the shoot and non-shoot buds on the three young apple tree varieties using the visible/near-infrared spectrometer. The spectrometer separated the wavelengths into six ranges, seven days before bud burst. The six ranges together with two components were analyzed for the 'Jonagold', 'Miyabi Fuji' and 'Orin'. The PC1 between 640-652 nm was the highest for 'Jonagold', between 654-680 nm it was highest for 'Miyabi Fuji' and between 704-766 nm it was the highest for 'Orin' (99.9%, 99.7% and 99.6% respectively).

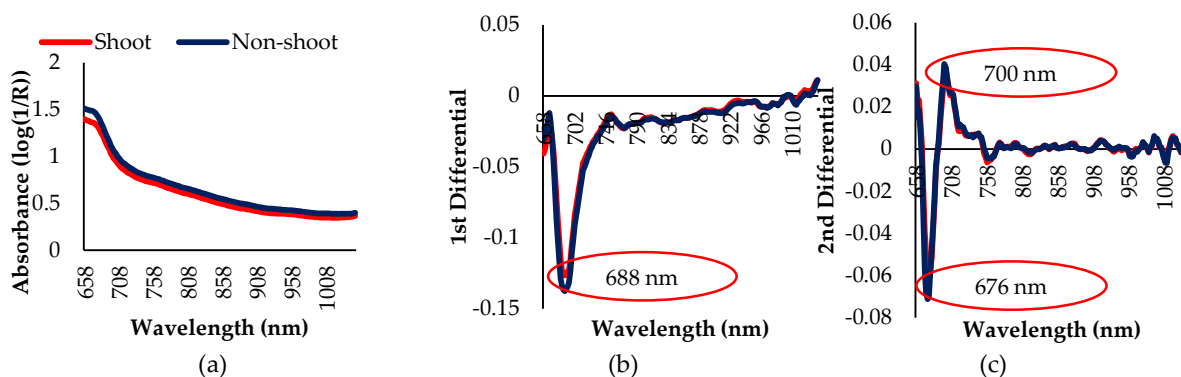


Figure-6. The light absorbance of the shoot and non-shoot buds for the 'Orin' seven days before bud burst. The 1st Differential and 2nd Differential on the visible/near-infrared spectrometer; (a) Absorbance (log(1/R)); (b) 1st Differential and (c) 2nd Differential.

Table-1. The PCA of the light absorbance of the shoot and non-shoot buds for the 'Jonagold', 'Miyabi Fuji' and 'Orin' trees. The wavelengths were separated into six ranges, seven days before the 2021 bud burst.

#	Separated wavelengths (nm)	Jonagold		Miyabi Fuji		Orin	
		PC1 (%)	PC2 (%)	PC1 (%)	PC2 (%)	PC1 (%)	PC2 (%)
1	640-652	99.9	0.1	99.5	0.5	63.1	30.9
2	654-680	99.7	0.2	99.7	0.2	98.8	1
3	682-702	98.2	1.8	98.3	1.7	97.4	2.6
4	704-766	99.6	0.4	99.5	0.5	99.6	0.4
5	768-924	98.7	1.2	99.5	0.5	99	1
6	926-1050	99.1	0.7	98.6	1.3	99.4	0.4

3.3. Classification analysis

Table 2 shows the classification analysis at 666 and 692 nm of the differences in light absorbance between the shoot and non-shoot buds using the data obtained from the visible/near-infrared spectrometer for 'Jonagold' on the days the buds were examined before the estimated date of bud burst. The accuracy, sensitivity and specificity of the classification for the 'Jonagold' shoot and non-shoot buds was analyzed. The highest percentage of accuracy for the distinguishing shoot from non-shoot buds one day before bud burst was near 76.6% with the Medium KNN of the KNN classifiers. The Medium Tree of the Tree classifiers had the highest sensitivity, about 75%, five days before bud

burst, while the specificity was 93% with the Medium Gaussian SVM of the SVM classifiers four days before bud burst.

Table-2. Classification analysis at 666 and 692 nm of the differences in light absorbance between the shoot and non-shoot buds using the data obtained from the visible/near-infrared spectrometer for the 'Jonagold' on the days the buds were examined before (May 6, 2021) the estimated date of bud burst; DBBB=days before bud burst.

Classifier	Classifier type	DBBB	Jonagold			Observation
			Accuracy (%)	Sensitivity (%)	Specificity (%)	
SVM	Linear SVM	VII	59.3	63	56	54
	Medium Gaussian SVM	VI	61.5	60	63	52
Tree	Medium Tree	V	68.1	75	63	47
SVM	Medium Gaussian SVM	IV	72.9	48	93	48
KNN	Weighted KNN	III	73.5	68	78	49
	Cosine KNN	II	70.2	70	70	47
	Medium KNN	I	76.6	75	77.8	47

Table 3 shows the classification analysis at 666 and 694 nm where the differences in light absorbance between the shoot and non-shoot buds using the data obtained from the visible/near-infrared spectrometer for 'Miyabi Fuji' on the days the buds were examined before the estimated date of bud burst. The accuracy, sensitivity and specificity of the classification for the 'Miyabi Fuji' shoot and non-shoot buds were analyzed. The highest percentage of accuracy for the distinguishing of shoot from non-shoot buds was near 82.1% with the Subspace KNN of the Ensemble classifier. The Quadratic SVM of the SVM classifier showed the highest percentage, about 85%, seven days before bud burst, while the specificity was 84% with the Coarse Gaussian SVM of the SVM classifiers one day before bud burst.

Table-3. Classification analysis at 666 and 694 nm of the differences in light absorbance between the shoot and non-shoot buds using the data obtained from the visible/near-infrared spectrometer for the 'Miyabi Fuji' on the days the buds were examined before (May 8, 2021) the estimated date of bud burst.

Classifier	Classifier type	DBBB	Miyabi Fuji			Observation
			Accuracy (%)	Sensitivity (%)	Specificity (%)	
SVM	Quadratic SVM	VII	80.7	85	75	57
	Quadratic SVM	VI	81.8	81	83	55
	Linear SVM	V	76.8	82	70	56

KNN	Cosine KNN	IV	78.6	76	83	56
Ensemble	Subspace KNN	III	82.1	81	83	56
SVM	Quadratic SVM	II	80.4	79	83	56
	Coarse Gaussian SVM	I	79.6	76	84	54

Table 4 shows the classification analysis at 666 and 694 nm of the differences in light absorbance between the shoot and non-shoot buds, using the data gained from the visible/near-infrared spectrometer for ‘Orin’ on the days the buds were examined before the estimated date of bud burst. The accuracy, sensitivity and specificity of the classification for the ‘Orin’ shoot and non-shoot buds was analyzed. The highest percentage of accuracy for the distinguishing of shoot from non-shoot buds one day before bud burst was near 76.3% with the Cubic SVM of the SVM classifier. The Cubic SVM of the SVM classifier had the highest sensitivity, about 76%, two days before bud burst, while the specificity was 77% with the Cubic SVM of the SVM two days before bud burst.

Table-4. Classification analysis at 666 and 694 nm of the differences in light absorbance between the shoot and non-shoot buds using the data obtained from the visible/near-infrared spectrometer for the ‘Orin’ on the days the buds were examined before (May 6, 2021) the estimated date of bud burst.

Classifier	Classifier type	DBBB	Orin			Observation
			Accuracy (%)	Sensitivity (%)	Specificity (%)	
Linear Discriminant	Linear Discriminant	VII	67.8	70	65	59
SVM	Medium Gaussian SVM	VI	65	68	62	60
	Medium Gaussian SVM	V	66.7	71	62	60
	Linear SVM	IV	74.1	59	42	58
KNN	Cubic KNN	III	71.2	70	73	59
SVM	Cubic SVM	II	76.3	76	77	59
Linear Discriminant	Linear Discriminant	I	64.4	61	69	59

4. Discussion

In Aomori Prefecture, young apple trees are usually transplanted in early spring. Before planting, these one-year-old trees are cut at the top, leaving 70-90 cm to form side branches, and the roots are shortened to about 10 cm for better rooting. This will create at least 3 to 5 side branches at the top of the young trees. Also, in these young trees, the process of bud change and pre-growth development occurs around 10-15 days after planting [1, 2, 9]. Determining the nature of the buds using a non-destructive device like the visible/near-infrared spectrometer is most appropriate shortly before bud burst for young trees.

The precision of the observations can be improved, initially, by detailing the internal development of the buds before bud burst, and likewise, by considering the entire annual

tree growth cycle as a single continuous process, where bud formation, stress induction, and bud vigilance spring issues identified to affect the timing according to Viherä-Aarnio et al. [12]. In this study, shoot and non-shoot bud light absorbance was tested using a visible/near-infrared spectrometer. We found that the light absorbance of the non-shoot buds was higher than that of the shoot buds for all measured dates and cultivars.

Additionally, there was little change in non-shoot bud light absorbance, although shoot bud light absorbance decreased near bud burst for both 'Miyabi Fuji' and 'Jonagold', no changes were observed for 'Orin'. The spectrometric data showed that the light absorbance of the shoot buds before bud burst was much lower than that of the non-shoot buds. The highest accuracy (82.1%) was observed with the Subspace KNN of the Ensemble classifiers test that was conducted on the "Miyabi Fuji" shoot and non-shoot buds three days before bud burst, whereas the highest (76.3%) was observed for 'Orin' buds two days before bud burst with the Cubic SVM of the SVM classifiers. However, seven days before bud burst, the highest (99.9%) first component of the PCA for 'Jonagold' was observed between 640-652 nm, while the lowest (99.6%) was observed for 'Orin' between 704-766 nm.

5. Conclusions

In conclusion, based on the above results, we intend to conduct a further study in open field conditions over a longer period of time. The results obtained in this study should help growers manage their orchards more precisely and at an earlier stage of tree growth. We suggest that a visible/near-infrared spectrometer be used to distinguish shoot buds from non-shoot buds before bud burst.

Author Contributions: For research articles with several authors, a short paragraph specifying their individual contributions must be provided. Conceptualization, methodology, software, validation, formal analysis, investigation and writing—original draft preparation, Alisher Botirov; data curation and supervision, Osamu Arakawa and Shuhuai Zhang. All authors have read and agreed to the published version of the manuscript.

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