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# **Image-based recognition using advanced neural networks can aid surveillance of *Agrilus* (Coleoptera, Buprestidae) jewel beetles**

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1 **Image-based recognition using advanced neural networks can aid surveillance of *Agrilus***  
2 **(Coleoptera, Buprestidae) jewel beetles**

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51 Running title: Automatic image-based identification of *Agrilus* species  
52

53 **Abstract**

54 The genus *Agrilus* includes two species, *A. planipennis* and *A. anxius*, that are of particular  
55 phytosanitary concern and that are regulated by the European Union legislation. This implies that  
56 phytosanitary agencies of all EU countries are obliged to establish specific surveillance programmes  
57 to verify the absence of these species from their territory. These activities commonly consist of the  
58 use of green-colored traps, which are however attractive not only for *A. planipennis* and *A. anxius* but  
59 also for a wide range of other *Agrilus* species. For this reason, much time and expertise is required to  
60 sort and identify specimens to species, impeding an efficient rapid response. In this study, we tested  
61 the efficacy of the Entomoscope, a low-cost, open-source photomicroscope that uses high-resolution  
62 digital imaging and allows a pre-trained CNN model to accurately detect, image and classify insect  
63 specimens, for automatic identification of 13 *Agrilus* species, including *A. planipennis* and *A. anxius*.  
64 The correct species was among the top five most probable predictions made by the trained CNN  
65 94.5% of times. For most species, including *A. planipennis* and *A. anxius*, either no errors or only a  
66 few errors were made, whereas for a few native species misidentifications were more common. The  
67 trained CNN also efficiently classified as “unknown” species that were not used in the training  
68 process. These results provided proof of concept for an AI-driven surveillance system that can  
69 strongly aid in surveillance activities of *Agrilus* species.

70  
71 **Keywords**

72  
73 *Agrilus planipennis*, *Agrilus anxius*, Bronze birch borer, Deep learning, Early-detection, Emerald ash  
74 borer, Entomoscope

## 75 **Introduction**

76 The constant increase of global trade over the last hundred years, combined with deliberate plant  
77 introductions in the past and ongoing climate changes, has facilitated the movement among continents  
78 and the establishment likelihood of an increasing number of insects (Brockerhoff and Liebhold 2017;  
79 Pureswaran et al. 2022; Fenn-Moltu et al. 2023; Isitt et al. 2024). The genus *Agrilus* (Coleoptera,  
80 Buprestidae), one of the most species-rich genera of animals in the world (Jendek and Grebennikov  
81 2023), is among the insect taxa that have taken the most advantage of these processes. More than 30  
82 species have been already introduced and established outside their native range (Ruzzier et al. 2023),  
83 including the emerald ash borer *A. planipennis* Fairmaire, 1888, which has caused massive ecological  
84 and economic damage in North America (Kovacs et al. 2010; Klooster et al. 2018). For these reasons,  
85 the development of tools and strategies for early detection of accidentally introduced *Agrilus* species  
86 was identified as a research priority to trigger rapid response and reduce potential impacts in the  
87 invaded areas.

88 Among the numerous tools developed for *Agrilus* surveillance programs, baited or unbaited  
89 green traps set up at entry points and in their surrounding forests are currently adopted by several  
90 phytosanitary agencies worldwide (Evans et al. 2020; Imrei et al. 2020; Silk et al. 2020; Dodds et al.  
91 2024; Duan et al. 2024; Santoiemma et al. 2024a). These traps were primarily developed based on  
92 lab and field studies targeting *A. planipennis* (Crook et al. 2009; Francese et al. 2010; Poland and  
93 McCullough 2014; Poland et al. 2019), but have been afterwards showed to be attractive also for a  
94 wide range of other species within the genus *Agrilus* (Rassati et al. 2019; Cavaletto et al. 2020; Le  
95 Souchu et al. 2024; Santoiemma et al. 2024a, Santoiemma et al. 2024b 2025; Khun et al. 2024). This  
96 allows practitioners to survey multiple species simultaneously, but much time and expertise is  
97 required to sort and identify specimens to species due to both the low interspecific morphological  
98 variation (Kelnarova et al. 2019) and the very high species richness of the genus (Jendek and  
99 Grebennikov 2023), impeding an efficient rapid response (Lyal and Miller 2020). Thus, novel  
100 technologies that can help to improve the efficiency of species identification are promptly needed.

101 Artificial intelligence (AI) systems are being increasingly adopted in entomology for various  
102 applications (Teixeira et al. 2023; Hartbauer 2024). Among them, AI is used to enhance the accuracy  
103 and speed of insect identification in the lab and in the field (Martinez et al. 2018; De Cesaro Júnior  
104 and Rieder 2020; Hartbauer 2024). Convolutional neural networks (CNNs) trained on image datasets  
105 have been shown to reliably classify insects at the family, genus or even species level (Valan et al.  
106 2019; Ärje et al. 2020; Hansen et al. 2020; Wührl et al. 2022; Tannous et al. 2023; Marais et al. 2024).  
107 Exploiting this technology in surveillance activities for *Agrilus* species would, however, require that  
108 laboratories and plant health inspectors are equipped with an affordable device that can capture  
109 images of trapped specimens and, subsequently, automatically identify them to species. The  
110 Entomoscope, recently developed by the Karlsruhe Institute of Technology (KIT) (Wührl et al. 2024),  
111 might satisfy these needs. It is a low-cost, open-source photomicroscope that uses high-resolution  
112 digital imaging and allows a pre-trained CNN to accurately detect, image and classify insect  
113 specimens (Wührl et al. 2024). The Entomoscope has been tested so far on parasitoid wasps (Shirali  
114 et al. 2024), but its potential application in *Agrilus* beetle surveillance has not yet been investigated.

115 In this study, we tested the efficacy of the Entomoscope and its associated CNN for automatic  
116 identification of *Agrilus* species collected in traps. We first trained the CNN using photos of 13  
117 *Agrilus* species taken with the Entomoscope, and then determined the trained CNN accuracy in  
118 identifying them to species. Second, we tested the response of the trained CNN to specimens  
119 belonging to species that were not included in the training process. Our objective was to provide proof  
120 of concept for the use of AI applied through the Entomoscope in surveillance activities targeting  
121 *Agrilus* species.

122

## 123 **Materials and Methods**

124

### 125 *Agrilus specimens*

126 Specimens used for the training process of the CNN belonged to 13 *Agrilus* species, eleven native to  
127 Europe and two non-native to Europe, i.e., *A. anxius* native to North America, and *A. planipennis*  
128 native to Asia but already occurring in Europe (Table 1, Fig. 1). The native species were selected as  
129 they were commonly collected in trapping studies carried out in Europe targeting *Agrilus* spp. (Le  
130 Souchu et al. 2024; Santoiemma et al. 2024b, 2025), whereas the non-natives were selected because  
131 they are regulated by the European Union legislation and surveillance is mandatory in EU countries  
132 (EFSA et al. 2020a, Santoiemma al. 2024b). Specimens were collected in Poland, France, Slovenia  
133 and Canada (Table 1) using green multi-funnel traps set up in the canopy of oak-dominated forests  
134 or poplar stands. Depending on the country, trap-collecting cups were filled with mixture (2:1) of  
135 propylene glycol and water (Slovenia), mixture (1:1) of ethylene glycol and water (Poland), mixture  
136 (2:1) of monopropylene glycol and water plus a drop of liquid dish detergent (France), a saturated  
137 solution of table salt in water plus a drop of liquid dish detergent to reduce surface tension (Canada),  
138 or kept dry but integrated with a section of mesh impregnated with  $\alpha$ -cypermethrin insecticide  
139 (Storanet®, BASF Pflanzenschutz Deutschland, Germany) (France). Specimens were preserved in  
140 ethanol until identification. Species-level taxonomic identifications were performed by Gianfranco  
141 Curletti, Eva Groznik, Maarten de Groot, Jerzy M. Gutowski, Alain Roques and Aurélien Sallé based  
142 on morphological traits, keys and other reference materials (Schaefer 1950; Farrugia 2007; Paiero et  
143 al. 2012).

144

### 145 *Imaging*

146 *Agrilus* specimens were photographed using the "plug-in" version of the Entomoscope, which  
147 requires connection to a computer with Microsoft's Windows operating system (Wühlrl et al. 2024,  
148 see Fig. S1 for a picture and detailed description of the hardware). Two Entomoscopes, provided by  
149 the KIT, were used. One was installed in the laboratory of the DAFNAE department at the University

150 of Padova, and the other was installed at the Entomology Museum of La Sapienza University of  
151 Roma. The software (ENIMAS) used for image acquisition is available free of charge from a GitLab  
152 repository (<https://gitlab.kit.edu/kit/iai/ber/enimas>) and Open Science Framework (OSF:  
153 <https://osf.io/3vmrg/> with DOI:10.17605/OSF.IO/3VMRG) (Wühlrl et al. 2024). The software allows  
154 one to choose between two photo stacking methods, one implemented in ENIMAS itself and the other  
155 one via a connection with Helicon Focus. For this study, we used Helicon Focus (version 8.2.2). For  
156 the species used to train the algorithm, we used a minimum of 11 to a maximum of 59 specimens per  
157 species, without distinguishing between sexes. Each specimen was photographed from multiple  
158 angles, corresponding to five main points of view: dorsal, laterodorsal, lateral, lateroventral and  
159 ventral (Fig. S2). This approach allowed us to obtain comprehensive coverage of a specimen's  
160 morphology. However, depending on the specimen and the position in which it was captured and  
161 preserved (the "death position"), it was not always possible to obtain images from all five angles as,  
162 in some cases, the death position did not allow us to hold the specimen at a precise angle. Therefore,  
163 two to five complete images were taken of each specimen. Specimens were not manipulated by  
164 taxonomists (or at least to a minimal extent for the identification process) but instead kept in the  
165 natural death position to simulate as much as possible the conditions faced by phytosanitary  
166 inspectors during surveillance programs. All specimens were photographed completely submerged in  
167 ethanol (70%) to avoid distortion effects that can cause significant identification errors.

168 Most images were obtained by stacking 18–25 photos, but as many as 35 photos were used  
169 for when specimens that had particular characteristics such as open elytra or outstretched legs. The  
170 distance between one picture and another was set at 0.07–0.1mm (depending on the size of the  
171 specimen) to obtain a homogeneous focus. The image acquisition speed was set between 10% to 15%;  
172 a higher speed proved counterproductive for alcohol-preserved specimens, due to the vibrations that  
173 caused the specimen to move. The images were then merged using the stacking function of Helicon  
174 focus (with respective settings: Method B; Radius 8; Smoothing 4), to obtain a single focused image  
175 for each angle of the specimen.

176

177 ***Training setup and model architecture***

178 The CNN was trained using a personal computer equipped with a NVIDIA RTX 4000 Ada Generation  
179 GPU, Intel Core i7-14700 2.10 GHz CPU and 32 GB RAM. Software: Python (v 3.10), PyTorch (v  
180 2.0.1), CUDA (v 11.7), Anaconda software (v 2.6.0) and Visual Studio Code (v 1.95.3). For  
181 automated image classification tasks, we used the You Only Look Once (YOLO) framework for  
182 classification, specifically YOLO11 (Jocher et al. 2023; Khanam and Hussain 2024). YOLO is a deep  
183 learning model designed to process entire images in a single forward pass, making it efficient for fast  
184 applications (Redmond et al. 2016, 2025). We implemented YOLO models through Ultralytics, a  
185 framework that provides streamlined tools for training, evaluation, and deployment of YOLO-based  
186 models. The YOLO models (like any supervised machine learning model) must be "trained", i.e., they  
187 must be provided with data, in this case images, that can allow the CNN to learn how to recognize a  
188 particular object on its own, without having to compare it with any other data (Limberg et al. 2022).  
189 Learning strongly depends on the quality and quantity of the training dataset: if the dataset well  
190 represents the variability of insects (e.g., different species, poses, and light conditions), YOLO can  
191 generalize better. The model can "learn" either specific patterns (e.g., stripes or spots) or that certain  
192 general shapes (e.g., elongated or round) belong to a certain class (Redmond et al. 2016). In our study,  
193 each class (a term used in the machine learning context and not as taxonomic rank) corresponded to  
194 a different *Agrilus* species. Convolutional filters can detect repeating and invariant patterns,  
195 regardless of rotations, illumination, or scale. YOLO divides each image into a grid and  
196 simultaneously predicts: i) the probability that a cell contains an object of a particular class (i.e.,  
197 species in our case); and ii) the parameters for the bounding box containing the object (Bochkovski  
198 et al. 2020). During the training, the models utilize various core configurations that influence the  
199 speed and the accuracy, such as learning rate, batch size and weight decay, but there are additional  
200 settings used to optimize the process, called argumentations. All YOLO argumentations are listed on  
201 the Ultralytics website at <https://docs.ultralytics.com/modes/train/>.

202 Images taken as described above were organized in three folders designated for CNN training,  
203 validation and testing (Table 1). For each species, 70% of the images were used for training, while  
204 the remaining 30% were equally divided into the folders designated for validation and testing.  
205 Although some species had more specimens than others, we decided to maintain the ratio 70:15:15  
206 for each species. The image dataset was curated following the guidelines provided in Ultralytics'  
207 documentation, which supports various datasets and labels specific to YOLO classification. The data  
208 was preprocessed by resizing the images to match YOLO's input dimensions, normalizing pixel  
209 values at 640x640, and augmenting images through transformations (i.e. flipud, fliplr, mosaic, mixup,  
210 etc., see Table S1).

211

### 212 ***Training procedure***

213 The training was conducted in Visual Studio Code (Microsoft 2024), an integrated development  
214 environment with Python (3.11.10) extensions, that allowed us to set up virtual environments and  
215 streamline package management for YOLO. Initially, all versions YOLO11 (i.e., *n*, *s*, *m*, *l* and *x*) were  
216 tested multiple times using a preliminary dataset with a reduced number of classes (i.e., fewer  
217 species). YOLO-*n* (nano) is the lightest, whereas YOLO-*x* (extra-large) is the most complex and  
218 accurate version but requires more computational resources. YOLO-*s*, *m*, and *l* fall between these two  
219 extremes, progressively increasing in complexity, number of parameters and accuracy at the cost of  
220 inference speed. In our case, the best-performing version was *x*. Consequently, only the *x* version was  
221 selected for the training procedure.

222 We used pre-trained models on ImageNet and fine-tuned them with training parameters that  
223 best suited our needs and our data. The training parameters, including batch size, learning rate and  
224 the number of epochs (where an epoch represents one complete pass of the training dataset through  
225 the algorithm) were defined as in Table S1. After training the model, its performance (i.e., the ability  
226 to assign a specimen to the correct species) was evaluated by analyzing the *results.csv* files generated  
227 in output directory, and by creating comparison charts. At this regard, two metrics of accuracy are

228 usually considered: Top-1 and Top-5. Top-1 is the percentage of times the model's prediction with  
229 the highest probability corresponds exactly to the correct class. When the model makes a prediction,  
230 only the first result (the one with the highest confidence) is shown; if it is correct, then it counts as a  
231 correct assignment. Top-1 is a very strict indicator of performance, because it requires that the most  
232 probable prediction is always the correct one. Top-5, on the other hand, is the percentage of times the  
233 correct class is among the top five most probable predictions made by the model. When the model  
234 makes a prediction, the first five classes to which the prediction is associated are shown. If the correct  
235 class is among the top five choices of the model, it is considered a correct prediction. This value is  
236 more useful for evaluating how close the model is to the correct identification, even if it does not  
237 always choose the right class first. For our study, we decided to use the Top-5 metric because it  
238 predicts the five most probable classes, which is an important feature for surveillance purposes. In  
239 fact, it allows us to know which native species the model tends to confuse with the targeted non-  
240 native species, allowing phytosanitary personnel to conduct subsequent additional morphological or  
241 molecular confirmations.

242 Besides Top-5 accuracy, we also calculated the F1-score. The F1-score is a performance  
243 measure of a classification model that combines *precision* and *recall* into a single value. *Precision*  
244 measures how many of the predicted positive cases are correct. *Recall* measures how many of the  
245 actual positive cases the model correctly identified. We also used *heatmap* (Kolde 2018) on RStudio  
246 to create a confusion matrix to graphically represent with a heatmap the accuracy of *Agrilus* species  
247 image classification and identification system.

248 Lastly, we used the Class Activation Mapping (CAM) technique (Zhou et al. 2016) in VSCode  
249 using EigenCAM (Muhammad and Yeasin 2020) to highlight class-specific regions of images that  
250 have more weight on the classification results. This technique superimposes a heatmap over the  
251 images of each specimen, producing images where the areas are colored on a scale ranging from red  
252 to blue, where red is applied to areas with more relevance for the classification, and blue is applied

253 to those with less (to no) relevance. Therefore, this can allow to spot the morphological characteristics  
254 or patterns that are used for the predictions.

255 All the images used for training and testing, as well as the scripts employed in the  
256 classification pipeline, are available in the Zenodo ([www.zenodo.org](http://www.zenodo.org)) repository at the following  
257 DOI: <https://doi.org/10.5281/zenodo.14998760>.

258

### 259 ***Testing the response of the CNN to species not included in the initial training***

260 To test the response of the trained CNN to specimens belonging to species that were not included in  
261 the training process, we added an additional class renamed "Unknown". In fact, YOLO does not  
262 include an "out-of-class" mechanism to exclude images that do not belong to any trained class, so it  
263 still tries to associate the image with the most similar species. When the image is very different from  
264 those used for training, the confidence level (max\_conf) of the prediction is typically low. In our case,  
265 we set this confidence level to 0.5, meaning that the model will ignore all predictions with less than  
266 50% confidence (Table S1). To test this mechanism, we used 19 images belonging to two native  
267 species that were not used for the training procedure (i.e., 8 images belonging to 2 specimens of *A.*  
268 *biguttatus* and 11 images belonging to 3 specimens of *A. convexicollis*, Table 1)

269

## 270 **Results**

### 271 *Identification accuracy of specimens belonging to species used in the training procedure*

272  
273 Trained CNN (i.e., YOLO11x) showed a high Top-5 accuracy (94.9%) in assigning specimens  
274 belonging to the *Agrilus* species used in the training process and the F1-score value was 77.2%. The  
275 trained CNN correctly assigned the highest percentage of correspondence to the correct species 100%  
276 of times for 6 species, i.e., the non-native *A. planipennis*, and the native *A. betuleti*, *A. cuprescens*, *A.*  
277 *graminis*, *A. obscuricollis*, and *A. olivicolor* (Fig. 2, Fig. S3). For three of the other species, the trained  
278 CNN assigned the highest percentage of correspondence to the correct species between 90.0% and  
279 95.0% of times (Fig. 2, Fig. S3). In particular, the native *A. angustulus* was identified with a 95%  
280 accuracy (two specimens out of 40 misidentified with *A. planipennis*), the native *A. laticornis* with a  
281 94.5% accuracy (one specimen out of 18 misidentified as *A. graminis*), and the non-native *A. anxius*  
282 with 90.0% accuracy (two specimens out of 20 were misidentified as *A. sulcicollis*). The *Agrilus*  
283 species for which a misidentification was common were *A. viridis*, *A. pratensis* and *A. hastulifer*. For  
284 *A. viridis*, the trained CNN assigned the highest percentage of correspondence to the wrong species  
285 (*A. graminis*) 16 out of 21 times (Fig. 2, Fig. S3). For *A. pratensis*, the trained CNN assigned the  
286 highest percentage of correspondence to the wrong species 11 out of 12 times, i.e., 9 times to *A.*  
287 *obscuricollis* and 2 times to *A. cuprescens* (Fig. 2, Fig. S3). For *A. hastulifer*, the trained CNN  
288 assigned the highest percentage of correspondence to the wrong species 15 out of 16 times, i.e., 12  
289 times to *A. graminis* and 3 times to *A. olivicolor* (Fig. 2, Fig. S3).

290 Considering class activation maps, to assign specimens to a certain species, the general shape  
291 of the body (pronotum and elytras) and the margins of the head (eyes and frons) were consistently  
292 used by the model (Fig. 3). Similarly, the shape of the abdominal sternites, the forehead, the area  
293 bearing the mouthparts (between clypeus, eyes margin and prosternum), and the clypeus itself were  
294 often used when the identification was based on pictures of the ventral part of the body.

295

### 296 *Identification accuracy of specimens not belonging to species used in the training procedure*

297 Considering specimens belonging to species not used in the training process, the trained CNN  
298 correctly assigned the highest percentage (100%) of correspondence to the class “Unknown” every  
299 time (Fig. 2, Fig. S3). This trend was valid both for the 8 specimens of *A. biguttatus* and 11 specimens  
300 of *A. convexicollis* (Fig. 2, Fig. S3).

301

## 302 Discussion

303  
304 The genus *Agrilus* includes two species, *A. planipennis* and *A. anxius*, that are of particular  
305 phytosanitary concern due to the economic and ecological impacts they can have in the invaded areas  
306 (Baranchikov et al. 2008, 2014; Kovacs et al. 2010; Klooster et al. 2018; Evans et al. 2020) and as  
307 such, they are regulated by the European Union legislation (EFSA et al. 2020a, EFSA et al. 2020b).  
308 This implies that phytosanitary agencies of all EU countries are obliged to establish specific  
309 surveillance programmes to verify the absence of these species from their territory (Evans et al. 2020).  
310 These activities commonly consist of the use of green-colored traps, which are however attractive not  
311 only for *A. planipennis* and *A. anxius* but also for a wide range of other *Agrilus* species (Rassati et al.  
312 2019; Cavaletto et al. 2020; Le Souchu et al. 2024; Santoiemma et al. 2024a, Santoiemma al. 2024b  
313 2025). Here we showed that the Entomoscope, in combination with the trained CNN, strongly aid in  
314 very properly discriminating *A. planipennis* and *A. anxius* from other commonly collected native  
315 *Agrilus* species, a process that demands much time and taxonomic expertise. In addition, we showed  
316 that the trained CNN can efficiently classify as “unknown” species that were not used in the training  
317 process, which is a key feature to efficiently alert phytosanitary personnel when a potentially non-  
318 native species other than *A. planipennis* and *A. anxius* has entered the country.

319 We showed that the CNN that we initially trained had a generally high precision in assigning  
320 a given specimen to one of the 13 *Agrilus* species on which the training process was based. The  
321 correct species was in fact among the top five most probable predictions made by the model 94.5%  
322 of times. On the one hand this is not surprising given that the CNN we used, i.e., YOLO models, were  
323 already demonstrated to be very useful for automatic insect classification (Takimoto et al. 2021; Stark  
324 et al. 2023; Gao et al. 2024), including woodborers (Zhang et al. 2024). On the other hand, the high  
325 precision that we obtained at the taxonomic level that we targeted (i.e., species) is less common (e.g.,  
326 Liu et al. 2023). For example, a study based on a previous version of the YOLO model showed a  
327 lower accuracy (i.e., 89%) in the identification of parasitoid wasps at the genus level (Shirali et al.  
328 2024). Overall, these results indicated that the CNN we trained starting from pictures taken with the

329 Entomoscope can be considered a very promising model to be used for the automatic identification  
330 of *Agrilus* species starting from individuals collected in traps.

331 We also found that the degree of accuracy with which the trained CNN assigned specimens  
332 to the correct *Agrilus* species varied among species. For most of them, including the non-native *A.*  
333 *planipennis* and *A. anxius*, either no errors or only a few errors were made, whereas for others (i.e.,  
334 the native *A. viridis*, *A. pratensis* and *A. hastulifer*) misidentifications were more common. This might  
335 be due to the characters used by the YOLO models to classify specimens. As for many others CNNs,  
336 YOLO models are only partially based on the same morphological characteristics (such as genitalia,  
337 hair distribution, size, etc.) (e.g., Volkovitsh et al. 2020) used by taxonomists but instead on the  
338 arrangement and intensity values in grayscale or Red-Green-Blue (RGB) of pixel clusters which are  
339 identified as discriminating features by the trained algorithm (Ganesh et al. 2021; Goodfellow et al.  
340 2016; Redmon et al. 2016; Bochkovskiy et al. 2020). Class activation maps showed, for example,  
341 that, depending on the position of the specimen, the general shape of the body and the margins of the  
342 head, elytras and pronotum or the shape of the abdominal sternites, the forehead and the clypeus were  
343 used as main characters to discriminate among the tested *Agrilus* species. This can result in a very  
344 accurate classification of species that are well known to be difficult to distinguish from congeners  
345 (i.e., *A. obscuricollis*), but also to failures when it comes to discriminate between species that can be  
346 instead easily separated using classic taxonomic approaches (e.g., *A. graminis* and *A. pratensis*)  
347 (Schaefer 1950; Farrugia 2007; Paiero et al. 2012). Overall, these results indicate that the  
348 Entomoscope and the current version of the trained CNN can be very useful for surveillance of *A.*  
349 *planipennis* and *A. anxius*, but are not yet ready to identify to species level all native species collected  
350 in traps.

351 Finally, we found that the trained CNN correctly assigned the highest percentage (100%) of  
352 correspondence to the class “Unknown” when trying to identify specimens belonging to species not  
353 used in the training process. Several trapping studies showed that several *Agrilus* species can be  
354 collected when using green-colored traps in forest areas surrounding entry points (e.g., Santoiemma

355 et al. 2024a, Santoiemma al. 2024b). These species might include native species present at low density  
356 or accidentally collected through the adopted trapping protocol, but also non-native species not  
357 initially targeted in the training process (e.g., non-regulated species). In this scenario, the capacity of  
358 the trained CNN to discriminate between known (i.e., species on which the training process was based  
359 on) and unknown species (i.e., species not included in the training process) can alert the phytosanitary  
360 personnel of the presence of a potential non-native species that should be further scrutinized via more  
361 classic morphological or molecular approaches. However, the chances that the “unknown” is a native  
362 species not included in the training of the CNN will increase with *Agrilus* species richness at the  
363 survey site; therefore, further efforts are necessary to increase the reference dataset with more native  
364 species.

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366

## 367 **Conclusions**

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369 By using the Entomoscope in combination with deep learning models we provide proof of concept  
370 for an AI-driven surveillance system capable of classifying the majority of *Agrilus* species with  
371 generally high accuracy. The open-source nature of this technology, combined with the relatively low  
372 cost of setting up and operating an Entomoscope, makes AI-based identification systems highly  
373 suitable for technology transfer initiatives. These systems could be integrated into training projects  
374 for phytosanitary personnel, foresters and environmental agencies, increasing knowledge on AI-based  
375 identification technologies, and encouraging their active participation in large-scale monitoring  
376 efforts. To further improve AI-based identification of *Agrilus* species, future research should explore  
377 the performance of other CNNs as well as hybrid approaches that integrate CNN-based image  
378 classification with other diagnostic methods, such as molecular tools. A multimodal approach,  
379 particularly in the initial training phases when large numbers of individuals need to be correctly  
380 assigned to their respective species, could significantly improve classification accuracy (Wuhrl et al.

381 2022). This would be particularly beneficial for species that are difficult to distinguish based on  
382 morphological characteristics alone. A major future advance would be the development of real-time  
383 processing capabilities, allowing field-based identification without the need for extensive laboratory  
384 infrastructure (Brydegaard et al. 2024; Chiavassa et al. 2024). A cloud-based or mobile application  
385 linked to the Entomoscope could allow users to upload images and receive automated identifications  
386 in real time, greatly increasing the accessibility and practical applicability of the system. Such an  
387 innovation would not only facilitate rapid species identification in phytosanitary surveillance but also  
388 contribute to broader biodiversity assessment initiatives.

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### 400 **CRedit authorship contribution statement**

401 **Valerio Caruso:** Writing - original draft, Methodology, Investigation, Formal analysis, Data  
402 curation, Conceptualization, Visualization; **Hossein Shirali:** Writing – review & editing,  
403 Methodology, Formal analysis; **Christophe Bouget:** Writing – review & editing; **Pierfilippo**  
404 **Cerretti:** Writing – original draft, Conceptualization, Supervision, Funding acquisition; **Gianfranco**  
405 **Curletti:** Writing – review & editing, Data curation; **Maarten de Groot:** Writing – review & editing,  
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412 Project administration

#### 413 **Declaration of competing interest**

414 The authors declare that they have no known competing financial interests or personal relationships  
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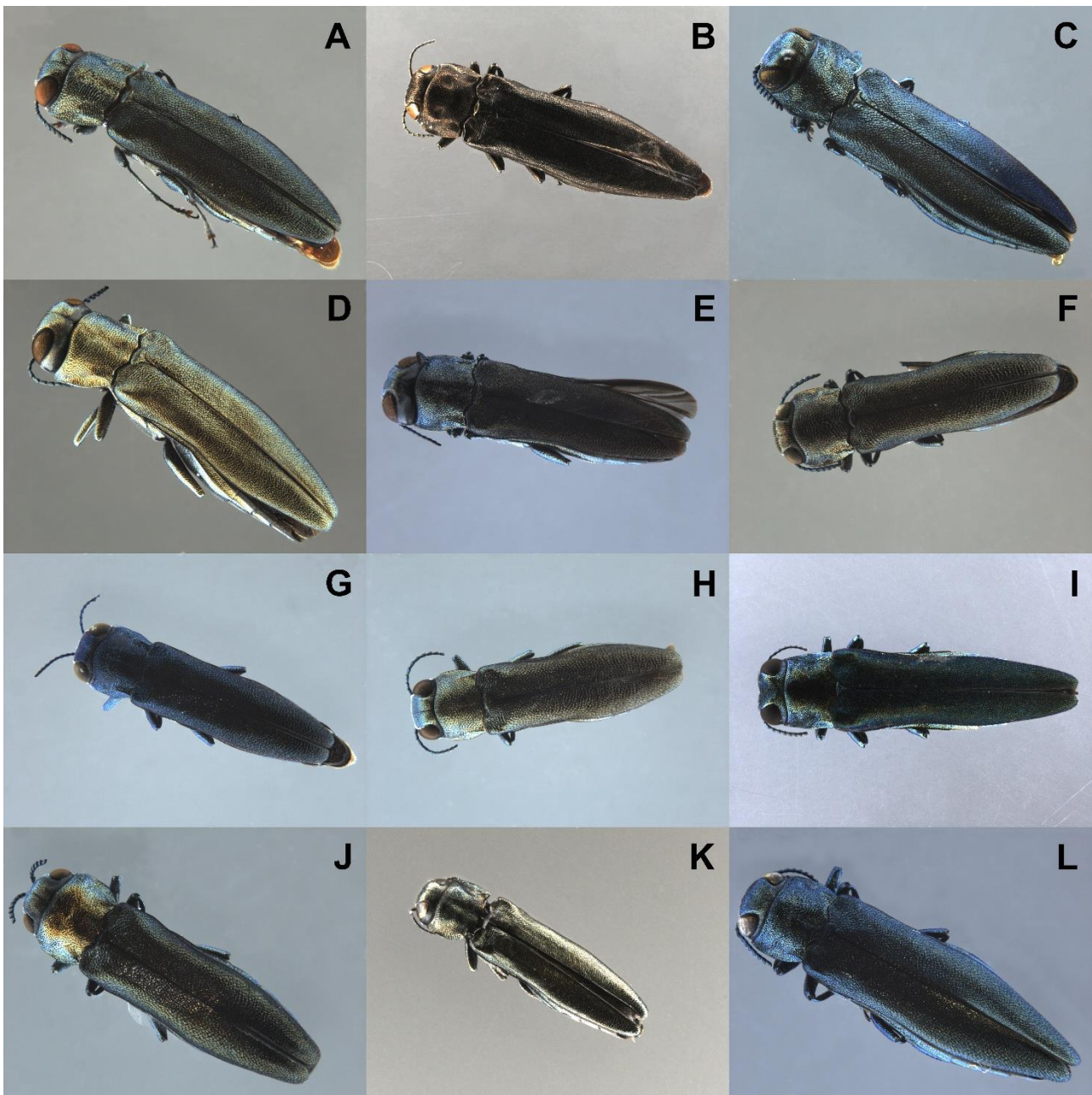
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678 **FIGURES**

679

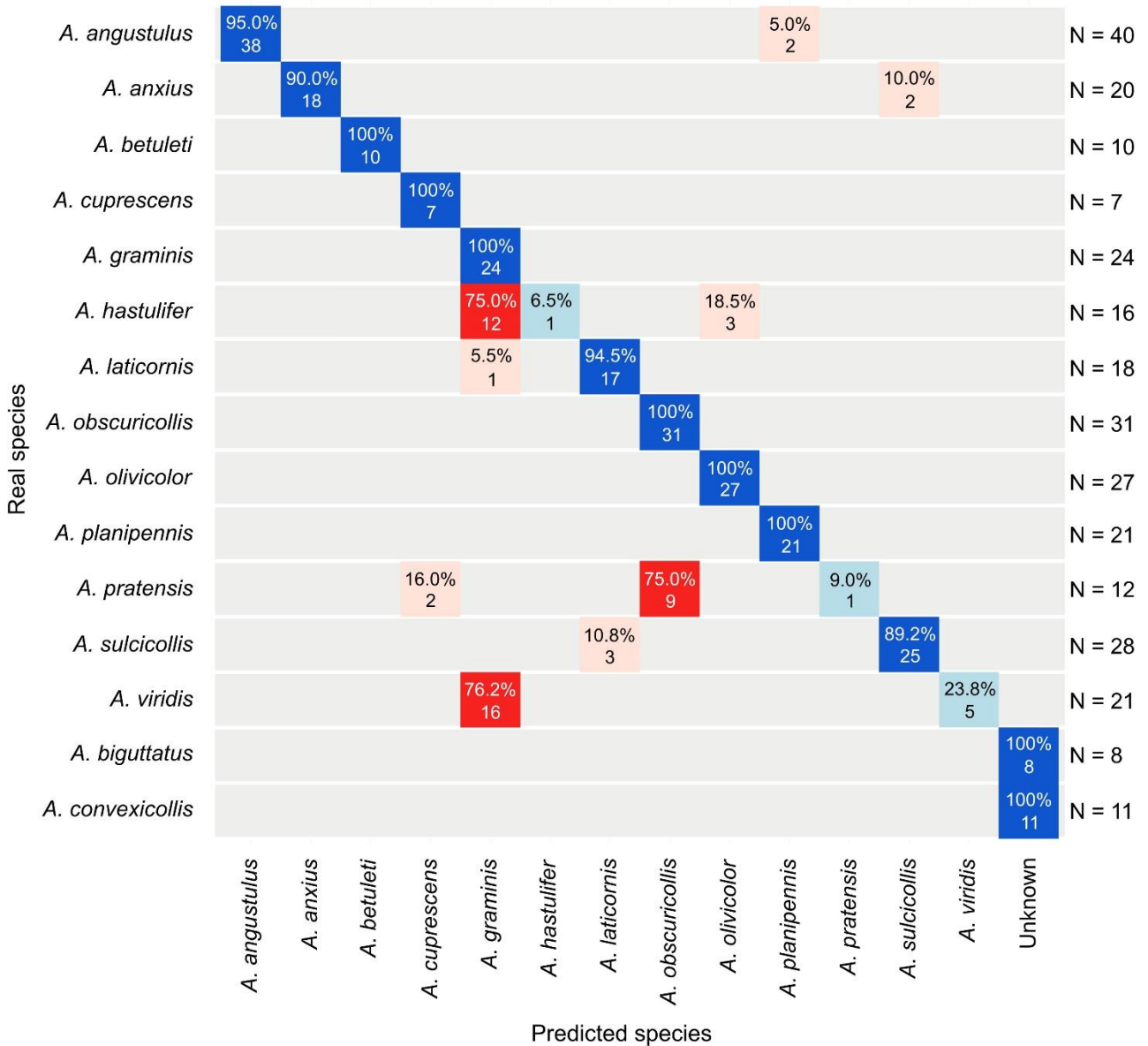
680 **Figure 1.** Examples of pictures taken using the Entomoscope to some of the *Agrilus* specimens  
681 used during the training process. (A) *A. angustulus*, (B) *A. anxius*, (C) *A. cuprescens*, (D) *A.*  
682 *graminis*, (E) *A. hastulifer*, (F) *A. laticornis*, (G) *A. obscuricollis*, (H) *A. olivicolor*, (I) *A.*  
683 *planipennis*, (J) *A. pratensis*, (K) *A. sulcicollis*, (L) *A. viridis*. Note: the pictures are not to scale  
684 with each other.  
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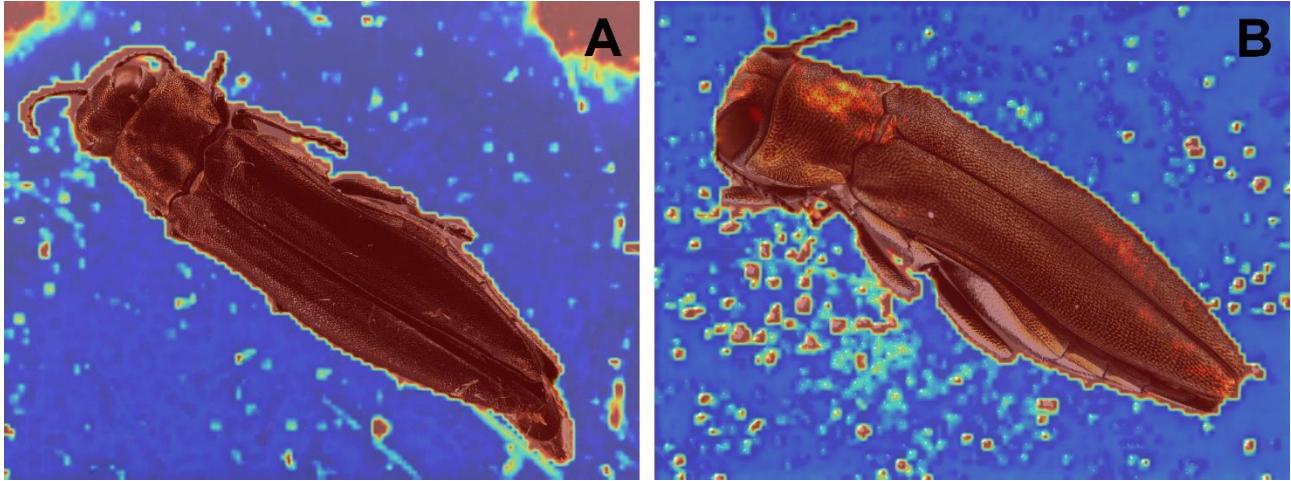
687

688 **Figure 2.** Confusion matrix with classification results of the YOLO11x model. Real species indicate  
 689 the true species identity. Predicted species indicate the species to which the model assigned analyzed  
 690 specimens. N = number of specimens for each species employed in testing process. The degree of  
 691 accuracy is reported both in the form of numerical values, showing both the percentage and the  
 692 absolute number of specimens that were assigned to a given species, and color-coded with blue for  
 693 correct predictions and red for incorrect ones (light blue and red are associated with low degree of  
 694 prediction, both correct and incorrect).  
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698 **Figure 3.** Example of class activation heatmaps obtained for pictures of A) *A. anxius* and B) *A.*  
699 *laticornis* (dorsal views). Colors range from blue to red based on the weight that areas had in  
700 classification: blue areas are regions with the least weight in classification, red areas are regions  
701 with the highest weight in classification, and the intensity of the red increases with the importance  
702 of the detected area.  
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706 **TABLES**

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708 **Table 1.** Number of specimens photographed and number of pictures used to train, validate and test  
 709 the CNN for each species. Countries where specimens of each species were collected are also  
 710 indicated. \*indicates species non-native to Europe. + indicates species used to test the response of the  
 711 trained CNN to species not included in the initial training process.

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Species	No. of specimens	No. of pictures for training	No. of pictures for validation	No. of pictures for testing	Country of collection
<i>Agrilus angustulus</i> (Illiger)	59	189	40	40	France, Poland, Slovenia
<i>Agrilus anxius</i> Gory *	35	96	20	20	Canada
<i>Agrilus betuleti</i> (Ratzeburg)	16	46	10	10	Poland
<i>Agrilus cuprescens</i> (Ménétries)	11	36	7	7	Poland
<i>Agrilus graminis</i> Kiesenwetter	46	115	24	24	France, Poland
<i>Agrilus hastulifer</i> (Ratzeburg)	28	74	16	16	France, Poland
<i>Agrilus laticornis</i> (Illiger)	36	85	18	18	France, Slovenia
<i>Agrilus obscuricollis</i> Kiesenwetter	47	144	31	31	France, Poland, Slovenia
<i>Agrilus olivicolor</i> Kiesenwetter	44	124	27	27	France, Slovenia
<i>Agrilus planipennis</i> Fairmaire *	34	95	21	21	Canada
<i>Agrilus pratensis</i> (Ratzeburg)	21	55	12	12	France, Poland
<i>Agrilus sulcicollis</i> Lacordaire	47	130	28	28	France, Poland, Slovenia
<i>Agrilus viridis</i> (Linnaeus)	40	98	21	21	Poland
<i>Agrilus biguttatus</i> (Fabricius) +	2	-	-	8	Slovenia
<i>Agrilus convexicollis</i> Redtenbacher +	3	-	-	11	France

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