Abstract
Traditional monitoring (e.g. in-water based surveys) of eelgrass meadows and perennial macroalgae in coastal areas is time and labor intensive, requires extensive equipment, and the collected data has a low temporal resolution. Further, divers and Remotely Operated Vehicles (ROVs) have a low spatial extent that cover small fractions of full systems. The inherent heterogeneity of eelgrass meadows and macroalgae assemblages in these coastal systems makes interpolation and extrapolation of observations complicated and as such, methods to collect data on larger spatial scales whilst retaining high spatial resolution is required to guide management. Recently, the utilization of Unoccupied Aerial Vehicles (UAVs) has gained popularity in ecological sciences due to their ability to rapidly collect large amounts of area-based and georeferenced data, making it possible to monitor the spatial extent and status of SAV communities with limited equipment requirements compared to ROVs or diver surveys. This paper is focused on the increased value provided by UAV-based, data collection (visual/Red Green Blue imagery) and Object Based Image Analysis for gaining an improved understanding of eelgrass recovery. It is demonstrated that delineation and classification of two species of SAV (Fucus vesiculosus and Zostera marina) is possible; with an error matrix indicating 86-92% accuracy. Classified maps also highlighted the increasing biomass and areal coverage of F. vesiculosus as a potential stressor to eelgrass meadows. Further, authors derive a statistically significant conversion of percentage cover to biomass ($R^2 = 0.96$ for Fucus vesiculosus, $R^2 = 0.89$ for Zostera marina total biomass, and $R^2 = 0.94$ for AGB alone, $P < 0.001$). Results here provide an example of mapping cover and biomass of SAV and provide a tool to undertake spatio-temporal analyses to enhance the understanding of eelgrass ecosystem dynamics.

Keywords: UAV, remote sensing, marine biology, coastal monitoring, seagrass, macroalgae

I Introduction
Many studies have shown that seagrass habitats are declining worldwide due to anthropogenic disturbances, such as eutrophication, habitat disruption and fragmentation, water turbidity, pollutants and coastal development (e.g. Flindt et al., 1999; Haynes et al., 2000; Orth et al., 2006; Williams, 2007; Waycot et al., 2009; Short et al., 2011; Moksnes et al., 2018). Many coastal systems suffer from high nutrient loadings
coming from agricultural runoff and sewage water which hamper natural recovery of seagrasses due to growth and shading from epiphytes. In other systems former seagrass habitats are now occupied by macroalgae, hindering their recovery by shading and ballistic impact from drifting macroalgae (Valdemarsen et al., 2010, 2011; Rasmussen et al., 2012; Canal-Vergés et al., 2014). Within Danish waters 40 years of historical eutrophication has induced excess sediment organic content such that the sediment anchoring capacity (the ability of the sediment to hold seagrass) also has been lost (Valdemarsen et al., 2014; Canal-Vergés et al., 2016; Flindt et al., 2016; Kuusemäe et al., 2016). Seagrasses are introduced as a “biological quality element in the European Union Water Framework Directive (WFD) for coastal and transitional waters” (WFD, CIS Guidance Document No. 20, 2009) due to the numerous ecosystem services they provide. These include, e.g., coastal protection and stabilization of the sediment (Ward et al. 1984), nutrient immobilization (Flindt et al., 1999), habitat formation and primary production (Boström & Bonsdorff, 2000) and carbon sequestration (Hemminga & Duarte, 2000). The objectives for the status of seagrasses in European and transitional waters are described in the WFD, annex V; “changes in angiosperm abundance due to anthropogenic activities should not be detectable” and “the levels of angiosperm abundance should be consistent with undisturbed conditions”.

In Denmark the depth limit of the seagrass species Zostera marina (eelgrass) is implemented as one of three biological quality elements and it is routinely measured (bi-yearly) along limited, 2-meter-wide transects located in estuaries and along the open coast (Bruhn et al., 2017). These efforts produce information about the growth conditions related to the light climate at the specific location of sampling, however, they do not provide spatial information about eelgrass coverage and production (e.g., the transects only represents permilles of the coastal system with no information of bed extent). Spatial information about eelgrass coverage is essential to understand how eelgrass recovery occurs in ecosystems that are highly altered when compared to the reference state (Orth et al., 2012; Benson et al., 2013). Only by including knowledge of full system coverage, and area-specific production is it possible to evaluate the state of ecosystem services provided by recovering eelgrass meadows. The complex and heavily heterogeneous spatial landscape structures in these systems, including macroalgal assemblages, eelgrass patches, and their varying coverage
all influence the ecosystem services provided by benthic vegetation. As such, collecting data with traditional techniques based on observations, photos and video recorded along narrow transects is time and labor intensive and requires considerable effort. The activity fulfills the aim of monitoring the depth limit of eelgrass, but it is not possible to use these observations to represent coverages of eelgrass at a landscape level, as the heterogeneity of these ecosystems hinders inter- and extrapolation between the transects.

Due to historical reduction in eelgrass distribution and biomass, a higher proportion of nutrients is available for macroalgae production in Odense Fjord (Valdemarsen et al., 2010). The macroalgae stocks, especially the highly mobile bladderwrack, Fucus vesiculosus, impact eelgrass beds by a range of stress-effects including deteriorated light environment, erosion and resuspension (Flindt et al., 2007; Canal-Vergés et al., 2009; 2013). Mobile macroalgae also has a destructive ballistic impact on eelgrass seedlings and mature beds when they are dragged by water currents while being attached to bigger stones (Canal-Vergés et al., 2010, Canal-Vergés et al., 2014). Here, F. vesiculosus physically damages seedlings as evidenced by high seedling mortality during periods of intense macroalgae drift (Valdemarsen et al., 2010). Further, when F. vesiculosus enters mature eelgrass patches, it can create additional fragmentation along the patch edges and internal bare areas as quantified by field observations (Flindt et al. 1997, 1999). Given the complex interactions between F. vesiculosus and eelgrasses, tools are required to rapidly and accurately assess the spatial extent and biomass of each SAV to inform management of areas where the two species co-occur.

Studies have shown that traditional satellite- and plane-based remote sensing of SAV has its limitations (Hossein et al. 2015) and that no methodology is suitable for assessing all relevant ecological parameters: presence/absence, cover, species, biomass and thus change. Due to the variable flight height, and hence resulting image resolution, of UAVs and the general Very High-Resolution (VHR) of UAV-imagery, it is argued that this platform is most suitable for covering more or all these parameters. Especially if the aerial imagery is combined with field measurements of SAV biomass, a complete overview of the ecological state and development can be obtained. In recent years UAV applications in marine areas have been increasingly utilized: Barrell and Grant (2015) presented a scheme for extracting information of eelgrass and blue mussels from imagery collected using a balloon. Duffy et al. (2018) presented a scheme for analysis of a seagrass and
surrounding macroalgae in the intertidal zone. Svane et al. (2016) presented a method for UAV-based mapping of eelgrass and macroalgae in the subtidal zone, and Nahirnick et al. (2019) outlined the benefits and problems in UAV-based mapping of seagrass in coastal zones. With the rapid development of UAV technology and associated software to support image interpretations by segmentation and classification of SAV based on spectra, shape and context (in short Object Based Image Analyses (OBIA)) (Blaschke, 2010), it is logical for ecological managers to attempt to integrate these new technologies with the data collected from more traditional methods (e.g., video sleigh or diver surveys). UAV-based, georeferenced, image-acquisition covering large areas, makes it possible to perform classifications across large spatial scales in comparatively small timescales (Joyce et al., 2017). By collecting images from high altitudes, which allows imagery collected to cover larger areas at the cost of a reduction in resolution, eelgrass patch structures and sizes may be characterized. Further, by using UAV-based imagery from low altitudes, UAV systems provide the possibility to assess the density of a patch; which in turn can be used as an indicator of local stressors to eelgrass. Duffy et al. (2018) also states the possibility of complementing spatial information collected (e.g., seagrass and macroalgae coverage) with biomass information to support the growing interest in carbon sequestration studies (e.g Mcleod et al. 2011; Howard et al. 2017; Sousa et al. 2019). This focus point also has relevance in relation to determining the plant bound nutrient transport which impacts estuarine areas (Salomonsen et al. 1999, Flindt at al. 2004, 2007).

The aim of this study was therefore to develop and test UAV-based analytical methods to 1) quantify eelgrass and macroalgae coverage, 2) obtain area-specific biomass estimates of eelgrass and *F. vesiculosus*, 3) monitor and quantify vegetative growth of eelgrass patches over time and 4) utilize fragmentation metrics as an indicator of the degree of stress present.

**II Methods**

**1 Study area**

Odense Fjord is a shallow estuary with an average depth of 2.2 m impacted by freshwater inputs from River Odense. Depending on freshwater input and exchange with Kattegat Sound, salinity varies from 5 to 25 ppm.
Petersen et al. 2009). Odense Fjord has a large catchment area (1046 km²) resulting in substantial nutrient loading primarily due to agricultural runoff. Prior to 1990 the nutrient loading was relatively high (2500 and 300 t N and P yr⁻¹) but after the implementation of several water action plans the nutrient loading was reduced to present levels (1500–2000 t N yr⁻¹ and 50 t P yr⁻¹) (Petersen et al., 2009). Reductions in nutrient loadings have improved the overall water quality, as evidenced by a reduction of phytoplankton concentrations and reduced blooms of opportunistic macroalgae (Petersen et al., 2009). However, Odense Fjord does not meet the targets set out by the WFD regarding eelgrass (Zostera marina) depth limits due to the current nutrient levels. In the reference condition, based on coverage data from 1880-1930, eelgrass covered substantial areas (> 15.1 km²) of Odense Fjord at Enebærodde (Petersen et al., 2009), while the coverage today is below 1.2 km². The environmental depth target in the EU WFD Water Management Plan is 4.2 m in Odense Fjord, corresponding to 75% eelgrass coverage in the estuary in the reference state. The eelgrass population has not shown signs of recovery – either in shallow or deeper areas of Odense Fjord – suggesting that light availability is only one of the many pressures affecting the current eelgrass distribution in the system. Valdemarsen (2010) demonstrated that physical stress from water currents and wave action together with ballistic impact from *F. vesiculosus* drifting along the seabed attached to stones hinder the survival of eelgrass seedlings and hence the natural recovery of eelgrass.

For this study two focus areas in the Northern Part of Odense Fjord were chosen for SAV mapping and analysis (Figure 1): Site (A) a nursery area for *F. vesiculosus* in the relatively protected North-West corner of Odense Fjord with a large monospecific standing stock of *F. vesiculosus* (approximately 400 ha). Site (B) contains a medium sized (0.6 hectare) eelgrass population with a high degree of fragmentation and a high-level of physical impact (Valdemarsen et al. 2010; Canal-Vergés et al. 2014). This region is situated in the Northern part of Odense Fjord with a water depth of 0.5 – 2 meters and is heavily exposed to wind and wave forces resulting from prevailing South Westerly winds (Danish Meteorological Institute, 2016). From site (A) large amounts of *F. vesiculosus* drift to site (B) where it is thought to hinder eelgrass recovery processes, with eelgrass patches continuously fragmented and never able to close the gaps and grow together.
2 Technical setup

The UAV platforms used in this project were consumer grade quadcopters with no sensor upgrades. The quadcopter type was chosen to be able to fly in alternating altitudes and perform UAV-based ground truthing but still being able to cover large areas. The UAV-model used for mapping *F. vesiculosus* at site B was a DJI Phantom 3 Professional (P3) with the standard 1/2.3” CMOS sensor with 12.4 million effective pixels and a field of view of 94° at a 20 mm f. 2.8 aperture (DJI, 2017). For all mapping at site A, a DJI Phantom 4 Professional (P4) with the standard 1” CMOS sensor with 20 million effective pixels and a field of view of 84° at an 8.8 mm/24 mm f. 2.8 – 11 aperture was used (DJI, 2017). These platforms were chosen as they are both accessible, cost-effective, and durable platforms with a high-quality camera. High capacity 4S LiPo (lithium-polymer) batteries of 5350-5870 mAh were used providing a practical flight time of 22-28 minutes depending on the wind conditions. For security reasons, no battery was used below the 20 % mark before return-to-land. This made mapping up to 15 hectares per battery possible when flying in 100 meters altitude.

3 Field procedure

**Biomass study:** To determine the relationship between the top-down surface area visible from aerial images and the biomass of *F. vesiculosus*, 15 samples of complete specimens were collected at site A (593725.58 m E; 6153864.01 m N) in July 2015 (*Figure 1*). The samples were collected randomly at different water depths (0.4 to 1.5 m) with samples exhibiting a variety of underwater shapes. The algal material was collected as single individuals and brought back to the laboratory. Here the specimens were carefully cleaned with demineralized water and epifauna, epiphytes and debris were removed. They were then oven-dried at 60 °C until constant weight. For each sample the wet weight and dry weight was measured.

To determine the relationship between coverage, shoot number and Above Ground Biomass (AGB) of eelgrass, samples were collected at 16 randomly chosen plots at site B (*Figure 1*) in September 2016. Sampling involved placing a 25x25 cm metal frame in the chosen plots and collecting all eelgrass biomass
inside the frame. Plant material were then carefully cleaned and processed as described for macroalgae above. The dry weight of the material was then determined and calculated per square meter. The POC (particulate organic carbon), C:N ratio and thus nutrient content was calculated from conversions outlined in the published literature (Kristensen, 1990; Duarte, 1990). At both sites the *F. vesiculosus* specimens and eelgrass sampling sites were overflown at 20- and 100-meter altitude in connection with the biomass sampling to be able to couple these. For all 100-meter flights it was assumed that eelgrass exhibited a percentage cover of 80% to standardize biomass calculations and prevent overestimation.

**UAV flight:** Based on preliminary test-flights a general mapping altitude of 100 meters corresponding to a Ground Sample Distance (GSD) of 3.8 cm (P3) and 2.6 cm (P4) was chosen. This provided a sufficient resolution for general classification on a landscape level. For studying classification efficiency of different coverages of eelgrass, flights in 20 meters altitude were performed. In each area, UAV-based ground truthing was tested and subsequently performed at 2-10 meters altitude (GSD: 0.05 – 0.38 cm) which was used for classification, validation and studies of classification limitations.

4 *Image analysis*

The production of georeferenced ortho-mosaics from single images was performed in Agisoft Photoscan ver. 1.3.4 and Agisoft Metashape ver. 1.5.5 (Agisoft, 2020). This photogrammetry software presents an easy-to-use workflow for stitching single images through steps covering correction, point cloud generation, surface construction, and orthomosaic production. All segmentation and classification of image objects was performed using the software Trimble eCognition Developer ver. 9.3 and 9.5 (Trimble Inc., 2020) by an OBIA approach (Lang, 2008; Blaschke, 2010) in which spectral information is combined with shape, distances, and context of pixels clustered into objects. All parameters in this analysis were determined by preceding testing to establish the optimal range for segmentation and recognition of the species of interest. For initial segmentation a multiresolution algorithm was used (Baatz & Schape, 2000) with the adjustable parameters: scale (100 m imagery = 70, 20 m imagery = 50), color/shape ratio (0.2), compactness (0.5) and equal weight of each color band (1,1,1). To secure an objective and comparable classification for different time-steps and between stations a supervised machine learning classification approach with training samples
was used (Lillesand, 2017). A Support Vector Machine (SVM) algorithm was chosen for the machine learning approach as it is precise (Tzotsos and Argialas, 2008) and less sensitive to low numbers of training samples than other machine learning algorithms (Myburgh & Van Niekerk, 2014). In all analyses a number of 20 samples per class was assessed as the optimum and applied for training. The object features used for classification were the mean and standard deviation of the spectral RGB (Red, Green, Blue) values of the pixels aggregated within each object and the mean difference to neighboring objects (distance = 0). The following classes were used in the classification: Macroalgae (here: *F. vesiculosus*) ‘Fv’, Eelgrass ‘Zm’, Bare bottom ‘BBx’ and the specific eelgrass coverage classes for low altitude mapping: Coverage of 1-50 % ‘Zm_SUB_50’, between 50 and 75 % ‘Zm_50_75’ and above 75 % ‘Zm_75_100’. Several classes of bare bottom were used and subsequently merged due to variance in their spectral features. A class covering solar glints was also introduced but did not have an impact on the analysis as this was performed on orthomosaics in which glints were removed.

For classification of *F. vesiculosus* at site (A) high altitude (100 m) orthophotos from September 2015 were used. For classification of eelgrass and *F. vesiculosus* at site (B) high altitude (100 m) orthophotos created from imagery acquired in the growth season 2015-2017 were used, and for detailed eelgrass coverage percentage classification at site (B) low altitude (20 m) orthomosaics created from imagery acquired in the growth season of 2016 were used.

**Accuracy assessment:** To assess the accuracy of classifications a confusion matrix (Foody, 2008; Congalton & Green, 2009) was constructed using the built-in accuracy assessment module in eCognition. Following the initial classification 10 subsets for each classification type were chosen in a stratified random manner for Fv (100 m) at site (A), Zm and Fv (100 m) at site (B), and low altitude Zm, Fv (20 m) at site (B). The classification accuracy was then determined from 100 (in 100 m imagery) and 50 (in 20 m imagery) validation points for each class chosen by stratified random sampling based on very low altitude imagery (< 2 m).
5 Statistical procedures

Relationships between UAV-based observations and biomasses of both *F. vesiculosus* and eelgrass together with eelgrass shoot density, was determined with Pearson’s linear correlation analysis. Data was tested for normal distribution (Shapiro-Wilk’s test) prior to the correlation analysis. All statistical procedures were performed in SigmaPlot v. 12 (SSI, 2019) using a significance level $p < 0.05$.

III Results

Results are divided into delineation and classification - including accuracy assessment – of the monitored organisms and objects of interest, biomass calculations of *F.vesiculosus* and eelgrass along with a combination of the two sections.

1 Delineation and classification

Both eelgrass and *F. vesiculosus* were delineated and classified in this study using the described methods. In the *F. vesiculosus*-dominated site (A), it was possible to extract the coverage (as summed top-down surface area) of the *F. vesiculosus* (Fv) along with separating individual and accumulated macroalgae from the bare bottom (BB) (*Figure 2*).

[Insert figure 2]

For the *F. vesiculosus* study at site (A) three classes were used: macroalgae denoted ‘Fv’ (*F. vesiculosus*), bare bottom 1 (BB1) and bare bottom 2 (BB2). As the sediment surface had varying spectral properties in the project area, due to varying organic content and/or coverage of benthic diatoms, two sediment-classes were needed. Using this delineation, it was possible to determine the coverage and total area of *F. vesiculosus*. Further, when combined with dry weight measurements (outlined below), it was possible to predict the biomass of *F. vesiculosus* in the project area (*Table 1*).

For the monitored eelgrass-beds at site (B), it was possible to delineate and classify objects of interest at varying levels of detail depending on flight altitude. For this study-area, low altitude (10 m flight height) a
total of 7 classes were used. It was possible to distinguish between different coverage classes and separate eelgrass from both bare bottom and *F. vesiculosus* (*Figure 3*).

[Insert figure 3]

All patch edges are classified as 1-50 % coverage as expected and even small variations in degree coverage were detected.

For determining eelgrass and *F. vesiculosus* general coverage and biomass on a large scale using high altitude images obtained at 100 m, four classes were used: Eelgrass (*Zm*), *F.vesiculosus* (*Fv*) and two classes of bare bottom (BB1 and BB2) (*Figure 4*).

[Insert figure 4]

Analyses showed a strong ability to delineate between eelgrass and bare bottom, including even macroalgae at the edges of the eelgrass beds. In zones with low coverage of eelgrass or macroalgae, misclassifications occurred (*Figure 4*).

2 **Accuracy assessment**

For all analyses performed on the varying flight heights and species an accuracy assessment was made (*Table 1*). An n = 10 and 50-100 validation samples per class were used for all classification types to secure a valid foundation for the assessment.

[Insert table 1]

The accuracy for classification of high-altitude imagery at site (A) and (B) was 94 ± 1 % and 93 ± 4 % with KIA (Kappa Indices of Agreement) values of 92 ± 3 % and 92 ± 3 % respectively, the latter expressing the probability of agreement with expected values (Congalton & Green, 2009). The low altitude imagery (site (B) 20 m) was > 86 ± 1 % expressed as overall accuracy and 83 ± 3 % expressed as KIA. At site (A) the largest source of error was *F. vesiculosus* being omitted (producer accuracy of 0.92 ± 0.03) and bare bottom being committed (user accuracy of 0.92 ± 0.02). For the high-altitude imagery at site (B) the largest source of
error was *F. vesiculosus* being omitted (producer accuracy of 0.83 ± 0.04) instead being classified as eelgrass. For the low-altitude imagery at site (B) the largest source of error was eelgrass with a 1-50 % coverage percentage being committed as either *F. vesiculosus* or eelgrass in with a higher coverage percentage (user accuracy of 0.65 ± 0.08). *F. vesiculosus* was equally being omitted (producer accuracy of 0.68 ± 0.03) due to being misclassified as low-coverage eelgrass or bare bottom.

3 Biomass study

It was possible to determine a direct and significant correlation between the surface area of bladderwrack (Fv) and its biomass in gDW (*Figure 5*) with a P-value < 0.001 an R²-value of 0.96.

[Insert figure 5]

The correlation shows low variation between the individual *F. vesiculosus* specimens with small individuals ranging 16-51 gDW to large individuals of 436-619 gDW biomass with surface areas ranging from 0.02 to 0.37 m².

It was equally possible to determine significant correlations for eelgrass. Significant correlations between percentage cover and shoot number along with degree of coverage and total biomass (AGB and BGB) in gDWm² were shown with P-values of < 0.001 and R² values of 0.95 and 0.89, respectively (*Figure 6*).

[Insert figure 6]

At this specific site a coverage degree of eelgrass of 25 % corresponded to a shoot number of 192 m⁻² and 95 % coverage to a shoot number of 1072 m⁻². When converting biomass (AGB only) to coverage a range from ~ 25 gDWm⁻² at < 10 % coverage to ~ 259 gDWm⁻² at > 95 % coverage could be measured. For Below Ground Biomass (BGB) the numbers ranged from ~ 10 gDWm⁻² at < 10 % coverage to ~ 646 gDWm⁻² at > 95 % coverage.
Using the classified images, it was possible to determine the zones where of highest physical impact. Further, once classifications were combined with the dry weight measurements it was then possible to calculate the total biomass of the eelgrass patches in the 2000 m² survey area (Fejl! Henvisningskilde ikke fundet.2).

[Insert table 2]

The analysis shows a clear growth of the eelgrass beds from 2015 to 2017, with the main patch growing from 279 to 334 m². This is an increase in biomass of 10 kg DW (AGB) assuming an eelgrass percentage cover of 80 % corresponding to 0.25 kg N and 0.038 kg P (Duarte, 1990). The P/A-ratio (perimeter/area) decreased nearly 10 % during the three years indicating decreasing complexity of the seagrass bed outer border.

Regarding the macroalgae in the focus area, the analysis provides a snapshot of the specific coverage and thus accumulated biomass which varies from 177 kg DW in 2016 to above 320 kg DW in 2015 and 2017 corresponding to 3.84 – 6.94 kg N and 0.34 – 0.62 kg P when reference nutrient values are coupled to the coverages (Kristensen, 1990; Lorenzo et al, 2017).

IV Discussion

**General findings:** The study illustrated the feasibility of using Very High Resolution (VHR) imagery for near coast mapping of submersed aquatic vegetation on both a landscape level at a high altitude and for ground truthing of individual species at lower altitudes. It was possible to achieve positive results for all the part-aims of this study. Delineation and thus determination of coverage of eelgrass and bladderwrack was achievable using the proposed methodologies and eCognition software for analysis. Compared to earlier studies (Barrell and Grant, 2015; Duffy et al, 2018) our study was performed in a subtidal area with constant water coverage which could pose issues (reflection, turbidity etc.) as described by Nahirmick et al in 2019. However, our study showed that the influences of these issues can be reduced by using a multi-feature approach made possible by using the OBIA-framework in which more reliance on shape and context is introduced. This finding is consistent with other literature, for example, (Tzotsos et al, 2008) who showed a high accuracy of 86-92 % and generally demonstrated robustness towards changing environmental conditions when mapping using an OBIA approach. The largest source of error was separating 1-50 %
coverage percentage eelgrass from macroalgae and bare bottom in low-altitude imagery (20 m) but for generally separating eelgrass, bare bottom and *F. vesiculosus* in high-altitude imagery (100 m) high accuracies (< 90 %) was found.

**Ecosystem services:** Delineation and classification processes provided distinct classes consisting of bare bottom, macroalgae and several classes of varying eelgrass coverages making analysis of recovery and stress possible (fig. 5). Collection of spatial data on eelgrass and macroalgae coverage across space and time is essential to understand how eelgrass is recovering in ecosystems that are highly altered when compared to the ecosystems where eelgrass is the dominating benthic primary producer (Orth et al., 2012; Benson et al., 2013, Flindt et al. 2016, Kuusemäe et al. 2016). Only by including knowledge of full system coverage and area-specific production is it possible to evaluate the ecosystem services that eelgrass is providing with respect to self-protection, immobilizing nutrients during the growth season and improving the light climate by reducing resuspension events. In Figure 4., delineation and classification of the images verify the dynamic of drifting macroalgae in an eelgrass area within 2015-2017, where the area specific macroalgae density and geographic position of individuals varies between years. Inflowing *F. vesiculosus* is most likely responsible for the variable growth dynamics (e.g., increasing P/A ratio) of the eelgrass beds here due to its inherent stress-effect (Flindt et al., 2007; Canal-Verges et al., 2010; 2013) and the inability of new patches to develop from seedlings in the inner part of the system (Valdemarsen et al., 2010).

Regarding the composition of the eelgrass patches in the area, highly detailed delineation is important as this provides information about ecosystem services provided by the patches along with information of stress-related processes. Highly fragmented patches of low size and high abundance, for example, have value as microhabitats (Hirst & Atrill, 2008) but are also an indicator of high physical stress (Fonseca & Bell, 1998). As edge effects and gap dynamics play a role in supporting faunal biodiversity (especially in relation to arthropods) (Tanner, 2005; Warry et al., 2009) and overall community structure, (Bell et al., 1999) determining these is important in understanding ecosystem functioning. Using the methods outlined in this study, it will be possible to monitor the mortality of small, newly established patches, which is known to be high (Olesen & Sand-Jensen, 1994). From 2015 to 2017 an areal expansion of 20.1 % (7.9 – 11.3 % y⁻¹) was
observed in the monitored area (Table 2). This rate is comparable to existing studies showing 19 % y\(^{-1}\) in larger patches while smaller patches show a much higher growth rate (Olesen & Sand-Jensen, 1994). The lower yearly growth in the study area corresponds well with the higher exposure to physical stress in this site.

Having the ability to precisely estimate the standing stock of eelgrass and macroalgae biomass and thus nutrient content is important both in defining the value of ecosystem services and monitoring seagrass restoration success (Lange et al, in prep.; Lange et al in prep.; Orth et al., 2020); to calculate the effectiveness of these habitats to sequester blue carbon (Howard et al, 2017; Sousa et al, 2018; Mazarrasa, 2018), and for validating ecological models (Kuusemäe et al, 2016). Macroalgae also have a somewhat undisclosed role as carbon sinks and as such precise biomass mapping may support future studies aiming to quantify this role (Krause-Jensen & Duarte, 2016).

Seagrasses are essential to achieve stable and well-working shallow estuarine and coastal ecosystems due to their numerous ecosystem functions and services (Orth et al. 2012, Flindt et al. 2016). Most of these services are associated to growth related processes, where increasing shoot, rhizome and root densities in existing patches, but also expansion into bare bottom areas creates more seagrass biomass. Increases in biomass immobilizes equivalent masses of dissolved inorganic nutrients (DIN and DIP) taken up by the plant for growth purposes. By combining UAV and field sampling, it was possible to achieve highly statistically significant correlations between surface areas of *F. vesiculosus* (R\(^2\) = 0.96 and P < 0.001) and eelgrass (R\(^2\) = 0.94 and 0.89, P < 0.001) and area specific biomasses. For *F. vesiculosus* the correlation was seemingly unaffected by the varying underwater shape of the specimens. The measured biomass of *F. vesiculosus* was between 600 and 700 gDW for specimens of app. 0.4 m\(^2\) in size which correspond well with known values of 800-2000 gDW m\(^{-2}\) at 80-100 % coverage (Öberg, 2006).

For eelgrass the measured AGB of 250 gDW m\(^{-2}\) and total biomass of 900 gDW m\(^{-2}\) at 100 % coverage (app. shoot density of 1300 m\(^{-2}\)) corresponds well with other studies (200-400 gDW m\(^{-2}\)) and total biomass of 700-800 gDW m\(^{-2}\) (Olesen and Sand-Jensen, 1994; Olesen, 1999). As we are focusing on one defined area with constant sediment conditions the biomass shown is the total of both AGB and BGB as the relationship
between the two biomass types is expected to be stable as well. The composition of the sediment, especially regarding organic content, plays a role in not only anchoring capacity but also eelgrass rhizome growth (Wicks et al, 2009), thus for studies in different areas unique relationships for ABG and BGB should be made. At Enebærodde (site (B), Figure 1) the organic content is relatively low (0.5 – 0.9 % LOI) (Valdemarsen et al, 2010). Using known values of POC, C:N and P (Kristensen, 1990; Duarte, 1990; Lorenzo et al. 2017) both the plant bound nutrients and the potential as nutrient sinks can be calculated for the two species.

These findings support using image derived estimation of biomass, nutrient content, and temporal change for future studies along with calculations of ecosystem services where improved eelgrass coverage supports less resuspension and a higher benthic light intensity (Orth et al. 2012). It is well-known that eelgrass coverage-to-biomass ratios vary depending on leaf length (Carstensen et al. 2016). Populations growing in areas with suboptimal light intensities (higher turbidity, deeper growing populations) usually produce longer leaves. It is therefore recommended to check the coverage-to-biomass ratio between sites to ensure that trends used are applicable to site-specific growth forms.

**Imagery for ground truthing:** Traditional ground truthing and validation of classification accuracy is based on on-ground sampling and often additional species-specific information (shoot length, percentage cover etc.) is also collected (Husson et al, 2016; Duffy et al, 2018; Nahirnick et al 2019). When identifying and validating species presence we argue that very-low altitude, high-resolution UAV-based images are sufficient as a method of ground truthing, as long as the water depth, turbidity and wave action is not disruptive of a clear image. We found that this approach could be used in approximately 90 % of the survey situations making it a fast and easy alternative to manual ground truthing. However, this is an active area of discussion and care should be taken as described by Congalton & Green, 2009. Alternatively, drone-based underwater ground truthing can be performed by either lowering a camera underneath the surface (Stæhr et al, 2019) or landing on the surface (Svane et al., 2018). A common trait of traditional ground truthing methods is their relatively high time demand compared to in-air ground truthing.
**Implementation:** Regarding implementation of the proposed methodology two steps are time consuming and need attention by the user: Collection of samples for classification based on ground truthing images, and collection of samples for validation. These steps are possible to automate in a higher degree by automatically labeling the class in ground truth images and creating a training point from the image meta data for location, but this is a work in progress. Generally, the rulesets used are universally applicable both in time and space, as long as training and validation are repeated for each site/time. The machine learning algorithm is based on spectral and context features which when applied on objects obtain a spatial dimension. Due to an approach where, neighboring objects are compared in a certain feature space, the specific environmental conditions on the time of image acquisition minimally impact the resulting classification. Regarding the timeframe for UAV-based monitoring, a thorough evaluation of Danish weather data from 2013 to 2015 was made in another project (Svane, unpub.). This showed that mapping would be possible in 69-75 % of the daytime not taking turbidity into account. Windy conditions also show a limited impact on recognition of eelgrass and *F. vesiculosus* specifically within Danish coastal waters (Svane, unpub.).

Compared to traditional monitoring methodologies - e.g., diver and ROV (Bruhn et al, 2017) - the UAV-based methodology differs in important aspects: The coverage of eelgrass or macroalgae is area-based for the whole study area instead of being based on points in a narrow transect, and the data processing is less time consuming as it can be highly automated. Compared to aerial and satellite-based (Frederiksen et al, 2004; Hogrefe et al, 2014; DHI, 2019) monitoring of SAV the UAV-based method provides a higher spatial resolution in a trade off with spatial extent. The improved spatial resolution (0.5 - 5 cm/pix) makes more accurate species delineation and classification possible and is thus a prerequisite for precise monitoring efforts. The method is equally suited for training and/or validation for national monitoring projects based on satellite imagery such as the Danish Aquatic Vegetation Map from 2019 (DHI, 2019) or if plane-based aerial imagery is used (Ørberg et al, 2018). Very High Resolution (VHR) UAV-based imagery with a GSD of 2-3 cm/pixel will in this case support the differentiation between species such as perennial macroalgae and seagrasses along with ‘dark’ zones such as mussel beds or channels.
Due to the ability to distinguish between vegetation types and at the same time perform both UAV-based ground truthing services and covering areas of up to 500-800 ha day$^{-1}$ per UAV we propose the method as a valid alternative or support to traditional SAV-mapping. As the majority of eelgrass populations inhabit coastal zones with low water levels the total area of interest is also limited, thus the traditional diver and ROV surveys will only be necessary for the deepest growth zones where UAV-camera spectra are unlikely to create distinguishable differences (Hansen, 2018).

For future studies we recommend further development of machine learning rulesets. This includes a shift towards more automated analyses in which neural networks and self-learning will be an important part. For intertidal studies a multispectral NIR-setup is highly recommended as the additional bands will strengthen classification accuracy (e.g. Barillé et al, 2010) while it has little use in subtidal settings due to the rapid absorption of light in the NIR-spectrum (Cho and Lu, 2010). Preliminary studies (Svane, unpub.) here show a distinct increase in delineation accuracy when using NIR-imagery of eelgrass beds with a maximum of 10-20 cm water coverage, limiting the practical use of the NIR-bands in large scale coastal mapping.

### V Conclusion

Using UAVs for marine monitoring in the subtidal zone using the proposed methodology is a feasible way of assessing both the classified species/object of interest, percentage cover, density, and biomass of SAV and thus the degree of potential stress from, in this case, drifting macroalgae on vulnerable species such as eelgrass based on common fragmentation parameters (e.g., perimeter-to-area ratio). Methods here may be useful tool for identifying areas suitable for restoration efforts and, by combining areas with specific biomass samples, it is also possible to assess the overall amount of nutrients and carbon contained in a local ecosystem along with local transport. We therefore propose that this method is implemented on a broader scale in national surveys and monitoring as it heightens the data quality and information level available and can thus make the basis for decision making more qualitative. In the future, specific stress-assessments and estimation of biomass-to-area relationships specific to more SAV-species and sites should be implemented to gain a better understanding of the fine scale landscape processes affecting shallow marine environments.
Finally, this new technique seems to have potential to optimize site selection for eelgrass restoration activities and provide a fast and easy-to-use tool for coastal environmental mapping (Lange et al., 2020, unpub.).

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