Original Article

Utility of dual-energy computed tomography in the association of COVID-19 pneumonia severity

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Aim: Coronavirus disease 2019 pneumonia differs from ordinary pneumonia in that it is associated with lesions that reduce pulmonary perfusion. Dual-energy computed tomography is well suited to elucidate the etiology of coronavirus disease 2019 pneumonia, because it highlights changes in organ blood flow. In this study, we investigated whether dual-energy computed tomography could be used to determine the severity of coronavirus disease 2019 pneumonia.

Methods: Patients who were diagnosed with coronavirus disease 2019 pneumonia, admitted to our hospital, and underwent dual-energy computed tomography were included in this study. Dual-energy computed tomography findings, plane computed tomography findings, disease severity, laboratory data, and clinical features were compared between two groups: a critical group (18 patients) and a non-critical group (30 patients).

Results: The dual-energy computed tomography results indicated that the percentage of flow loss was significantly higher in the critical group compared with the non-critical group ($P < 0.001$). Additionally, our data demonstrated that thrombotic risk was associated with differences in clinical characteristics ($P = 0.018$). Receiver operating characteristic analysis revealed that the percentage of flow loss, evaluated using dual-energy computed tomography, could predict severity in the critical group with 100% sensitivity and 77% specificity. However, there were no significant differences in the receiver operating characteristic values for dual-energy computed tomography and plane computed tomography.

Conclusion: Dual-energy computed tomography can be used to associate the severity of coronavirus disease 2019 pneumonia with high accuracy. Further studies are needed to draw definitive conclusions.

Key words: COVID-19 pneumonia, COVID-19 pneumonia severity, DECT, lung perfusion blood volume, plane CT

INTRODUCTION

CORONAVIRUS DISEASE 2019 (COVID-19) pneumonia causes progressive respiratory failure, despite the maintenance of lung compliance.[1] The pathophysiology of severe hypoxemia in COVID-19 pneumonia is gradually becoming clearer and several studies have now suggested that severe acute respiratory syndrome coronavirus 2 (SARS-CoV2) induces unique coagulation disorders that result in microvascular thrombosis in the lung.[2–5] Impaired pulmonary perfusion, associated with microthrombosis, is not a characteristic of ordinary pneumonia and results in a specific phenomenon called "happy hypoxia," in which patients do not experience respiratory distress despite hypoxemia.[6]

Lung scintigraphy is a common method of assessing pulmonary perfusion; however, it is usually associated with nuclear medicine and cannot be performed as an urgent test.[7] Dual-energy computed tomography (DECT) correlates with lung scintigraphy and allows for the assessment of pulmonary perfusion in a manner that is suitable for urgent examination.[8] DECT has been suggested to be effective in visualizing the etiology of COVID-19 pneumonia because of its ability to highlight changes in organ blood flow.[9,10]

Although plane computed tomography (CT) has been widely performed in the clinical management of COVID-19 pneumonia, DECT is not currently applied as a routine test.

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There are reports that lung perfusion blood volume, measured by DECT, is lower in intubated or poorly oxygenated patients and is associated with disease duration.[11,12] However, the clinical utility of DECT for COVID-19 pneumonia remains unknown.

SARS-CoV2 infection continues to be a global pandemic, causing severe illness, usually in the form of COVID-19 pneumonia. To maintain the medical system, the risk of severe disease should be evaluated, to better identify patients in need of hospitalization and transport to higher medical institutions.

Therefore, we designed this retrospective study to predict disease severity by evaluating and comparing the utility of DECT and plane CT in understanding the pathogenesis of COVID-19 pneumonia.

METHODS

Research design

THIS STUDY WAS a single-center, retrospective, observational study. Patients ages 16 years or older, admitted to our hospital with a diagnosis of COVID-19 pneumonia, between August 2020 and January 2021, with suspected pulmonary embolism, were included in the study. Patients who were unable to undergo contrast-enhanced CT on the discretion of their attending physician, because of renal dysfunction, allergy or other medical conditions, were excluded. The included patients were divided into two groups: a critical group that required high-flow nasal cannula or intubation during hospitalization and a non-critical group.[13] We then assessed and compared the percentage of blood flow defects using DECT, percentage of lesions using plane CT, laboratory data, such as lymphocyte count ($\times 10^6$/mL), C-reactive protein (mg/dL), aspartate aminotransferase (AST; U/L), lactate dehydrogenase (LDH; U/L), glomerular filtration rate (GFR) (mL/min), ferritin (ng/mL), Krebs von den Lungen (KL)-6 (U/mL), interleukin (IL)-6 (pg/mL), fibrin degradation products (FDP) (μg/mL), D-dimer (μg/mL), total plasminogen activator inhibitor (PAI)-1 (ng/mL), and clinical characteristics, such as age in year, sex as man and woman, onset to admission in days, existing thrombosis risk defined as history of cancer, chronic kidney disease, arterial disease, or cerebral infarction,[14,15] past history of diabetes mellitus, obesity, body mass index, smoking history, inhaled steroid, systemic steroid, steroid pulse, antiviral agent, anticoagulant therapy, length of oxygen therapy, length of hospital stay, length of ICU stay, oxygen therapy, mechanical ventilation, in-hospital death, ratio of peripheral blood oxygen saturation to fraction of inspired oxygen (SpO_2/FiO_2) for the patients in each group. A primary outcome was disease severity, which was classified as the critical or non-critical group.

DECT imaging protocol and image analysis

Computed tomography scans were obtained using a second-generation dual-source CT scanner (SOMATOM Definition Flash; Siemens Healthcare, Forchheim, Germany) in the dual-energy scan mode. These evaluations were completed using the following parameters: detector collimation, 32 × 0.6 mm; 285 ms rotation time; pitch, 0.55; and tube voltage, 140 kV with a tin (Sn) filter and 100 kV. The slice thickness and increment were set to 2.0 and 1.4 mm, respectively. A medium-soft convolution kernel with iterative reconstruction (Q33 iterative strength 2) was used to produce all the images.[10]

Plane CT was also taken during DECT imaging (Fig. 1A). The area of the whole lung field was pulled out from the plane CT (Fig. 1B). Next, the pneumonia lesion area as was extracted (Fig. 1C). DECT images were output in two different modes using SyngoVia software (Siemens Healthcare) in the lung perfused blood volume (PBV) mode and liver virtual non-contrast (VNC) mode. The lung PBV mode evaluates perfusion in the lungs and does not show areas other than optimal perfusion in these tissues (Fig. 1D). The default settings suggested by the manufacturer were used during analysis; therefore, a region of 0.5 cm^2 in each area was placed in the pulmonary trunk and used as the reference vessel to calibrate the color coding. The air density was set to −1,000 HU on both the 100 and 140 kV (Sn) images, and the soft tissue density was set to 57 HU on the 100 kV images and to 55 HU on the 140 kV (Sn) images. The contrast medium ratio, minimum border, maximum border, resolution, and contrast medium cutoff were set to 2.24, –960 HU, –600 HU, 4, and –50 HU, respectively.

The liver VNC mode allows for the concentration of the contrast agent to be freely set, with the algorithm generating a map encoding the iodine content in each CT voxel. Therefore, we used 100% contrast medium to identify any areas of increased pulmonary perfusion (Fig. 1E).

The lung PBV and liver VNC modes were then superimposed to delineate areas of reduced pulmonary perfusion because of COVID-19 pneumonia (Fig. 1F). These regions of decreased ventilatory blood flow were then identified as the lesion area for DECT. The lesion scores (percentage, %) for DECT and plane CT were calculated using the image analysis program Image J (NIH, Bethesda, MD).[16,17] Briefly, the CT data from three 5-mm slices evaluated using plane CT and the lung PBV and liver VNC images from the DECT imaging were imported into Image J. Image J then extracted the lesions in each slice using the fixed thresholds, and the DECT and plane CT scores were calculated for each slice using the following formulas before

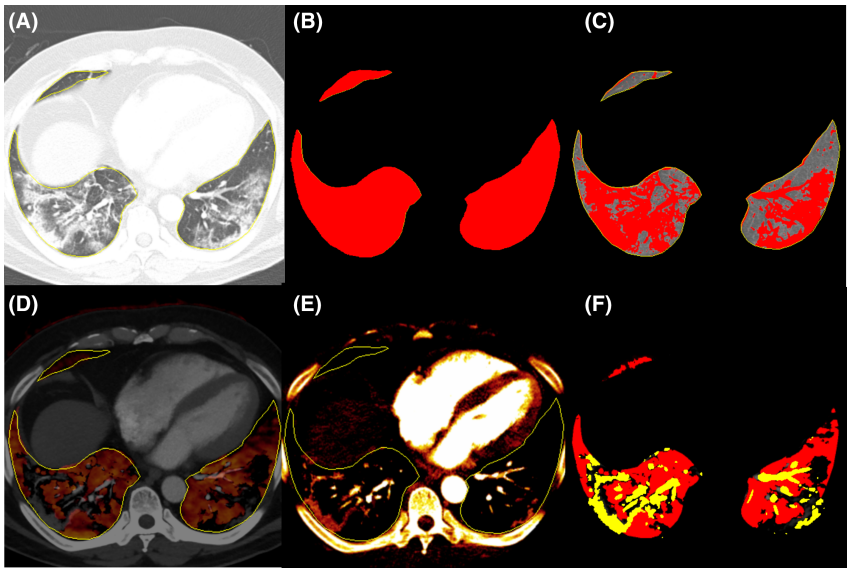

Fig. 1. CT image evaluation protocol. (A) Plane CT of a patient with COVID-19; (B) The area of the whole lung field when pulled out of the plane CT; (C) Extracted area from the white part of the lung field; (D) DECT evaluation of COVID-19 patient (lung PBV mode); (E) DECT evaluation of COVID-19 patient (liver VNC mode); (F) Area other than the area of decreased blood flow in the lung (overlapped area of D and E). COVID-19, coronavirus disease 2019; CT, computed tomography; DECT, dual-energy computed tomography; PBV, perfused blood volume; VNC, virtual non-contrast.

the results from each slice were combined (threshold values were evaluated and determined by two emergency physicians):

$$\text{DECT score} = 1\text{–}f/b \text{ and}$$

$$\text{Plane CT score} = c/b,$$

where b is the area of the lung field (Fig. 1B); c is the pneumonia lesion area on plane CT (Fig. 1C), and f is the area outside the area of decreased pulmonary perfusion on DECT (Fig. 1F).

STATISTICAL ANALYSIS

CATEGORICAL DATA WERE expressed as numbers (percentages) and continuous variables were expressed as medians (quartiles), unless otherwise stated. Categorical data were compared using Pearson's χ^2/Fisher's direct probability test, and continuous variables were compared using Student's *t*-test or Mann–Whitney *U* test based on the distribution of the data. A two-tailed P value of <0.05 was considered statistically significant. Receiver operating characteristic (ROC) analysis was performed to define the value of each of the parameters in association with disease severity and inclusion in the critical group. Next, we compared area under the curve (AUC)s of DECT scores, plane CT scores, SpO_2/FiO_2, with other study variables, using the DeLong's method. All statistical analyses were performed using EZR (Saitama Medical Center, Jichi Medical University, Saitama, Japan), a graphical user interface for R (R Foundation for Statistical Computing, Vienna, Austria).[18]

RESULTS

A TOTAL OF 79 patients met the initial inclusion criteria for our study. Thirty-one patients were excluded because of unsuitability, and 48 patients were included in the study (Fig. 2).

Table 1 summarizes the clinical characteristics of the 48 patients, divided into two groups: 18 in the critical and 30 in the noncritical group. The median age in both groups was 75 years (quartile, 62–81 years). A total of 38 males (79%) were enrolled in this study. The critical group comprised of only males; therefore, there was a significant difference in sex between the two groups ($P = 0.008$). There was no difference in smoking history, body mass index, or presence of diabetes mellitus between the two groups, but there was a significant difference in thrombotic risk ($P = 0.018$). When assessing variations in treatments of COVID-19 pneumonia commonly used at our hospital, only the incidence of steroid pulse was shown to be significantly different between these

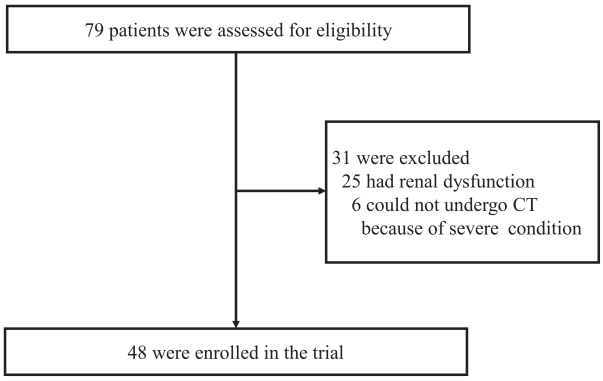

Fig. 2. Flowchart describing patient enrollment and exclusion. CT, computed tomography.

two groups ($P < 0.001$). There were 10 intubated patients (21%) and 7 deaths (15%) within this cohort, and the median plane CT score was 35% (quartile, 24%–47%). The median DECT score was 13% (quartile, 6.1%–22%) and the median SpO_2/FiO_2 was 290 (quartile, 230–440). All showed a significant difference between the two groups ($P < 0.001$).

No significant difference was observed with respect to the lymphocyte count, GFR, AST, KL-6, and IL-6 levels between these two groups (Table 2).

To identify patients in the critical group, ROC curves were evaluated (Table 3). The AUC of the DECT score was as high as 0.9, and the sensitivity and specificity of the ROC curve were 100% and 77%, respectively, with a cutoff of 13%. The plane CT scores were also highly accurate, with an AUC of 0.92. No significant difference was found between the ROCs for the DECT and plane CT scores (Fig. 3). In addition, SpO_2/FiO_2 associated the critical group with a sensitivity of 83% and specificity of 90% at a cutoff of 260 (Fig. 3).

The AUC of the laboratory data was lower across the board than the plane CT and DECT scores. The AUC of the

Table 1. Demographic and clinical characteristics of patients with COVID-19

	No. (%)			*P* value
	Total ($n = 48$)	Non-critical group ($n = 30$)	Critical group ($n = 18$)	
Age, median (IQR), year	75 (62–81)	73 (58–82)	77 (73–80)	0.265
Sex				
Female	10 (21)	10 (33)	0	0.008
Male	38 (79)	20 (67)	18 (100)	
Onset to admission, median (IQR), day	7.0 (4.0–11)	6.5 (4.0–11)	8.5 (6.0–12)	0.298
Risk of thrombosis[†]	21 (44)	9 (30)	12 (67)	0.018
Diabetes mellitus	21 (44)	12 (40)	9 (50)	0.558
Obesity	19 (40)	11 (37)	8 (44)	0.762
BMI (kg/m^2), median (IQR)	24 (22–26)	24 (22–27)	24 (22–27)	0.785
Smoking history	28/39 (72)	15/24 (63)	13/15 (87)	0.150
Inhaled steroid	44 (92)	28 (93)	16 (89)	0.624
Systemic steroid	33 (69)	22 (73)	11 (61)	0.522
Steroid pulse	15 (31)	2 (6.7)	13 (72)	<0.001
Antiviral agent	44 (92)	27 (90)	17 (94)	1.000
Anticoagulant therapy	46 (96)	29 (97)	17 (94)	1.000
Length of oxygen therapy, median (IQR), days	6.0 (3.0–15)	4.0 (2.0–6.0)	16 (11–27)	<0.001
Length of hospital stay, median (IQR), days	12 (10–19)	11 (7.3–13)	21 (14–31)	<0.001
Length of ICU stay, median (IQR), days	3.0 (0–10)	0 (0–3.0)	11 (7.3–14)	<0.001
Oxygen therapy	41 (85)	23 (77)	18 (100)	0.020
Mechanical ventilation	10 (21)	0	10 (56)	<0.001
In-hospital death	7 (15)	0	7 (39)	<0.001
DECT score, median (IQR)	0.13 (0.061–0.22)	0.08 (0.050–0.12)	0.22 (0.18–0.37)	<0.001
Plane CT score, median (IQR)	0.35 (0.24–0.47)	0.26 (0.20–0.34)	0.50 (0.44–0.59)	<0.001
SpO_2/FiO_2, median (IQR)	290 (230–440)	370 (290–450)	190 (14–250)	<0.001

BMI, body mass index; CT, computed tomography; DECT, dual-energy computed tomography; ICU, intensive care unit; IQR, interquartile range; SpO_2/FiO_2, ratio of peripheral blood oxygen saturation to fraction of inspired oxygen.
[†]Patients with one or more following conditions: history of cancer, chronic kidney disease, arterial disease, and/or cerebral infarction.

Table 2. Laboratory findings of patients with COVID-19

	Median (IQR)			*P* value
	Total ($n = 48$)	Non-critical group ($n = 30$)	Critical group ($n = 18$)	
Lymphocyte count ($\times 10^6$/mL)	800 (600–1,100)	860 (610–1,100)	720 (530–1,100)	0.217
CRP (mg/dL)	8.8 (4.3–13)	6.6 (2.2–11)	11 (8.9–21)	0.018
AST (U/L)	40 (27–59)	38 (26–48)	56 (42–69)	0.043
LDH (U/L)	310 (260–480)	290 (250–350)	490 (330–570)	0.002
GFR (mL/min)	64 (49–86)	66 (52–87)	59 (47–71)	0.310
Ferritin (ng/mL)	460 (270–840)	420 (240–520)	850 (510–1,100)	0.001
KL-6 (U/mL)	400 (250–560)	300 (240–490)	440 (330–800)	0.099
IL-6 (pg/mL)[†]	53 (24–110)	42 (24–72)	95 (30–130)	0.135
FDP (μg/mL)	3.4 (2.3–6.7)	2.7 (2.0–4.3)	4.9 (3.2–17)	0.033
D-dimer (μg/mL)	1.3 (0.6–3.3)	0.8 (0.5–1.9)	2.3 (0.9–7.1)	0.016
Total PAI-1 (ng/mL)[‡]	48 (33–67)	38 (31–61)	61 (50–88)	0.012

AST, aspartate aminotransferase; COVID-19, coronavirus disease 2019; CRP, C-reactive protein; FDP, fibrin degradation products; GFR, glomerular filtration rate; IL, interleukin; IQR, interquartile range; KL, Krebs von den Lungen; LDH, lactate dehydrogenase; PAI, plasminogen activator inhibitor.
[†]Non-critical group ($n = 27$), critical group ($n = 12$).
[‡]Non-critical group ($n = 26$), critical group ($n = 14$).

Table 3. Receiver operating characteristic analysis for the need of intensive treatment

	AUC	95% CI	Optimal cutoff	Sensitivity (%)	Specificity (%)
DECT score	0.90	0.82–0.99	0.13	100	77
Plane CT score	0.92	0.81–1.00	0.42	89	93
SpO_2/FiO_2[†]	0.89	0.77–1.00	260	83	90
Lymphocyte count ($\times 10^6$/mL)[†]	0.61	0.44–0.78	840	72	53
CRP (mg/dL)	0.71	0.55–0.87	8.8	78	67
AST (U/L)	0.68	0.50–0.86	41	78	67
LDH (U/L)	0.77	0.61–0.93	370	72	80
GFR (mL/min)[†]	0.60	0.43–0.77	72	78	47
Ferritin (ng/mL)	0.78	0.64–0.93	670	67	87
KL-6 (U/mL)	0.65	0.47–0.82	760	33	97
IL-6 (pg/mL)	0.64	0.45–0.84	56	69	70
FDP (μg/mL)	0.69	0.52–0.85	3.6	72	67
D-dimer (μg/mL)	0.71	0.56–0.86	4.5	44	90
Total PAI-1 (ng/mL)	0.75	0.58–0.92	46	92	63

AST, aspartate aminotransferase; AUC, area under the curve; CI, confidence interval; CRP, C-reactive protein; CT, computed tomography; DECT, dual-energy computed tomography; FDP, fibrin degradation products; GFR, glomerular filtration rate; IL, interleukin; KL, Krebs von den Lungen; LDH, lactate dehydrogenase; PAI, plasminogen activator inhibitor; SpO2/FiO2, ratio of peripheral blood oxygen saturation to fraction of inspired oxygen.β
[†]Only SpO_2/FiO_2, lymphocyte count, and GFR are positive below cutoff.

plane CT score was significantly different from all of the laboratory parameters. However, the DECT score was not significantly different from LDH, ferritin, and total PAI-1 levels ($P = 0.07$, $P = 0.109$, and $P = 0.086$, respectively), but was significantly different from the other laboratory parameters (Table 4).

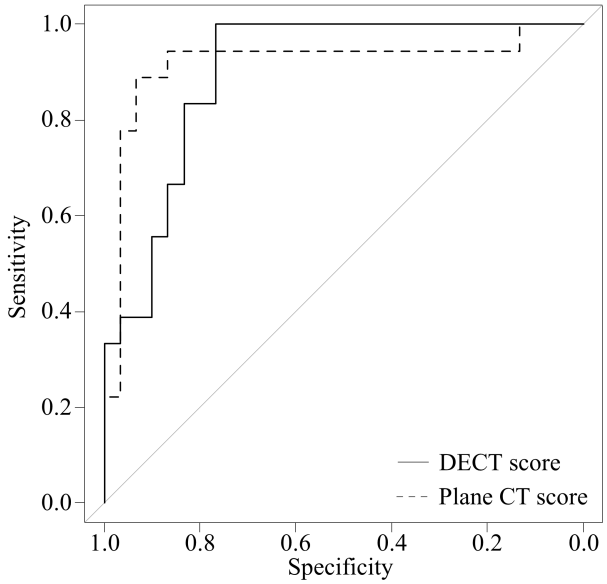

Fig. 3. ROC curve showing the DECT and plane CT scores when evaluating the need for intensive treatment. CT, computed tomography; DECT, dual-energy computed tomography; ROC, receiver operating characteristic.

Table 4. Comparison of receiver operating characteristic analysis for the need of intensive treatment (P-value)

	AUC	DECT score 0.90	Plane CT score 0.92	SpO_2/FiO_2 0.89
Lymphocyte count	0.61	0.001	0.001	0.003
CRP	0.71	0.024	0.002	0.026
AST	0.68	0.007	0.003	0.005
LDH	0.77	0.068	0.027	0.027
GFR	0.60	0.001	0.003	0.019
Ferritin	0.78	0.109	0.043	0.160
KL-6	0.65	0.008	0.006	0.005
IL-6	0.64	0.011	0.005	0.037
FDP	0.69	0.012	0.005	0.032
D-dimer	0.71	0.016	0.014	0.065
Total PAI-1	0.75	0.086	0.018	0.096

AST, aspartate aminotransferase; AUC, area under the curve; CRP, C-reactive protein; CT, computed tomography; DECT, dual-energy computed tomography; FDP, fibrin degradation products; GFR, glomerular filtration rate; IL, interleukin; KL, Krebs von den Lungen; LDH, lactate dehydrogenase; PAI, plasminogen activator inhibitor; SpO_2/FiO_2, ratio of peripheral blood oxygen saturation to fraction of inspired oxygen.

DISCUSSION

THIS STUDY REPORTED two major findings. First, DECT may be a significantly better predictor of severe disease in COVID-19 pneumonia than most laboratory tests. Among the numerous blood tests used in the clinical management of COVID-19, the most useful markers of severe disease include elevated leukocyte count, low lymphocyte count, and elevated D-dimer, C-reactive protein, LDH, ferritin, creatinine, KL-6, and IL-6 levels.[19,20] Here, we compared the lymphocyte count, DECT scores, and C-reactive protein, AST, LDH, GFR, ferritin, KL-6, IL-6, FDP, D-dimer, and total PAI-1 levels between the patient groups. The ROC curve for the laboratory parameters showed that the AUC for laboratory data was 0.6–0.783 for association inclusion in the critical group and 0.9 for the DECT score. The DECT score predicted severe disease with a sensitivity of 100% and specificity of 77%, at a cutoff of 13%, suggesting that DECT is more sensitive than other tests when screening patients with COVID-19 pneumonia.

Second, DECT and plane CT showed no difference in predicting the severity of COVID-19 pneumonia. Plane CT scores have been reported to correlate with the severity of COVID-19 pneumonia and are useful in predicting mortality.[21] Here, we compared the DECT scores with the plane CT scores of COVID-19 pneumonia patients, using ROC analysis. This ROC evaluation revealed that both scores exhibited a high degree of association value for identifying patients in the critical group, but there was no significant difference between DECT and plane CT. The DECT score assessed only the part of the lung where perfusion was reduced, whereas the plane CT score assessed the entire lung, including atelectasis and bacterial pneumonia. Because atelectasis and bacterial pneumonia are also involved in hypoxemia, the plane CT and DECT scores did not seem to differ in predicting disease severity.

Although we could not show that DECT is superior to plane CT in predicting the severity of COVID-19 pneumonia, DECT may be more useful in understanding the pathogenesis of COVID-19 pneumonia.

Because DECT evaluates pulmonary perfusion, it can visually diagnose atelectasis and secondary bacterial pneumonia, both difficult to distinguish on plane CT. Specifically, pulmonary perfusion is decreased in COVID-19 pneumonia and increased in atelectasis and bacterial pneumonia.[10,22] The perfusion distribution may help to determine whether the prone, right-sided, or left-sided position is more effective. Here, we present cases for which prone therapy was effective and non-effective. Effective cases showed a marked decrease in blood flow on the dorsal side of both lung fields, as detected by DECT. They were more likely to

benefit from the prone position than other patients. Therefore, continued prone therapy helped to improve oxygenation. In non-effective case, a diffuse marked decrease in blood flow, including on the ventral side, was detected by DECT; however, continued prone therapy showed no improvement in oxygenation (Fig. 4).

Prone therapy is effective in COVID-19 pneumonia and has been reported to significantly reduce mortality in intubated patients.[23] However, it is not clear which patients with COVID-19 pneumonia benefit from prone therapy.[24] Therefore, DECT may help determine the treatment strategy. This study only focused on the association of COVID-19 pneumonia severity using DECT, and further studies on the relationship between DECT and the understanding of the pathogenesis of COVID-19 pneumonia should be conducted.

This study had several limitations. First, this was a single-center study; therefore, the external validity of these findings is limited, and there is a possibility of selection bias. Second, because of the large number of exclusions, another selection bias may have occurred. The reason was that the cutoff value for renal function was not set, and it was the attending physician's decision whether to perform contrast-enhanced CT in patients with renal dysfunction. Third, the number of days from disease onset was not uniform as the CT imaging was performed immediately after hospitalization. COVID-19 pneumonia is prone to short-term changes in pathology, and the number of days from disease onset may have a significant effect on the imaging findings.[25]

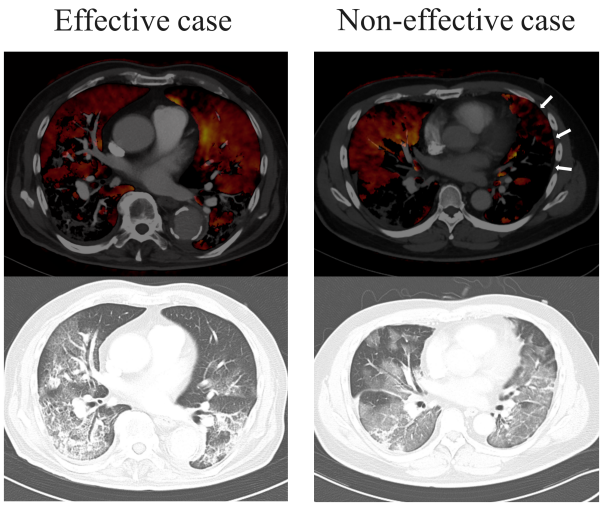

Fig. 4. DECT and plane CT images on admission of COVID-19 pneumonia patients for effective and non-effective prone position therapy. The effective case was a 79-year-old man. DECT showed a marked decrease in ventilation and blood flow on the dorsal side of both lung fields, suggesting that prone position therapy is effective. Prone position therapy improved oxygenation, from PaO_2/FiO_2 = 100 on day 1 to PaO_2/FiO_2 = 200 on day 6 of hospitalization. The non-effective case was a 58-year-old man. DECT showed a diffuse, marked decrease in blood flow, including on the ventral side (white arrow). The patient was on continued prone position therapy, but oxygenation did not improve and showed no improvement from PaO_2/FiO_2 = 91 on day 1 to PaO_2/FiO_2 = 89 on day 6 of hospitalization. COVID-19, coronavirus disease 2019; CT, computed tomography; DECT, dual-energy computed tomography; PaO_2/FiO_2, ratio of partial pressure of arterial oxygen to fraction of inspired oxygen.

CONCLUSIONS

DUAL-ENERGY COMPUTED TOMOGRAPHY can be used to predict the severity of COVID-19 pneumonia with high accuracy. However, in this study, DECT and plane CT showed no significant difference in predicting the severity of COVID-19 pneumonia. DECT may be more useful than plane CT for understanding the disease status of COVID-19 pneumonia, but further studies are needed to draw definitive conclusions.

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DISCLOSURE

APPROVAL OF THE Research Protocol: The Institutional Ethics Committee at the National Hospital Organization of the Yokohama Medical Center reviewed and approved the study protocol.

Informed Consent: This retrospective observational study used existing medical data, and it was difficult to obtain consent from patients for participation in this research. Information about this research was disclosed in an information disclosure document, and research subjects and others had the opportunity to refuse participation. If a refusal was offered, the data were not used. However, no refusals were offered in this study.

Registry and the Registration No. of the Study/Trial: Reception number 2020–40.

Animal Studies: Not applicable.

Conflict of Interest: None declared.

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