Markerless Video-Based Gait Analysis in People With Multiple Sclerosis

Matteo Moro, Giorgia Marchesi, Maria Cellerino, Giacomo Boffa, Francesca Odone, Matilde Inglese, and Maura Casadio

***Abstract*— Gait analysis plays a crucial role in assessing mobility impairments and monitoring disease progression in individuals with Multiple Sclerosis (MS). Markerless, video-based methods offer a non-invasive, practical alternative to traditional marker-based systems, making them particularly suitable for clinical applications. This study employs a markerless video-based approach to extract spatio-temporal and kinematic parameters from 25 individuals with MS and 25 age- and sex-matched unimpaired controls. The MS cohort was divided into two subgroups based on the Expanded Disability Status Scale (EDSS): "high" disability (*EDSS* $\geq$ 3) and "low" disability (*EDSS* < 3). Both normal and tandem gait patterns were evaluated. In normal gait, significant spatio-temporal and joint kinematic differences were observed between the high EDSS group and unimpaired controls, while the low EDSS group exhibited no notable deviations. In contrast, tandem gait analysis revealed significant differences in heel-to-toe distance between the low EDSS group and unimpaired controls, highlighting subtle changes that were undetectable in normal gait. These findings underscore the potential of video-based methods to enhance disease monitoring and guide targeted rehabilitation strategies in MS.**

Received 9 January 2025; revised 11 April 2025 and 9 June 2025; accepted 9 July 2025. Date of publication 16 July 2025; date of current version 22 July 2025. This work was supported in part by European Union—NextGenerationEU and the Ministry of University and Research (MUR), National Recovery and Resilience Plan (NRRP), Mission 4, Component 2, Investment 1.5, Project "RAISE—Robotics and AI for Socio-economic Empowerment" under Grant ECS00000035; and in part by the Fondazione Italiana Sclerosi Multipla under Grant FISM–2019/PRsingle050. *(Francesca Odone, Matilde Inglese, and Maura Casadio contributed equally to this work.) (Corresponding author: Matteo Moro.)*

This work involved human subjects or animals in its research. Approval of all ethical and experimental procedures and protocols was granted by the Institutional Ethical Committees (CER Liguria) under Approval No. 222REG2017 and the Department of Informatics, Bioengineering, Robotics, and Systems Engineering at the University of Genoa under Protocol No. CE DIBRIS-008/2020, and performed in line with the Declaration of Helsinki.

Matteo Moro and Francesca Odone are with the Department of Informatics, Bioengineering, Robotics and Systems Engineering (DIBRIS), University of Genova, 16145 Genova, Italy, and also with the Machine Learning Genoa (MaLGa) Center, 16146 Genova, Italy (e-mail: matteo.moro@unige.it; francesca.odone@unige.it).

Giorgia Marchesi and Maura Casadio are with the Department of Informatics, Bioengineering, Robotics and Systems Engineering (DIBRIS), University of Genova, 16145 Genova, Italy (e-mail: maura.casadio@unige.it).

Maria Cellerino, Giacomo Boffa, and Matilde Inglese are with the IRCCS San Martino Hospital, 16132 Genova, Italy, and also with the Department of Neurosciences, Rehabilitation, Ophthalmology, Genetics, Maternal and Child Health (DINOGMI), University of Genova, 16132 Genova, Italy (e-mail: maria.cellerino@unige.it; giacomo.boffa@unige.it; m.inglese@unige.it).

Digital Object Identifier 10.1109/TNSRE.2025.3589765

***Index Terms*— Gait analysis, video analysis, computer-aided motion analysis, multiple sclerosis.**

I. Introduction

GAIT analysis is a critical tool in the field of medicine and rehabilitation, enabling healthcare professionals to evaluate and monitor motion patterns in individuals with various conditions, including Multiple Sclerosis (MS) [9]. Multiple Sclerosis is a chronic neurological disease that affects the central nervous system, leading to impaired motor function and gait abnormalities [16]. Therefore, analyzing gait in people with MS is important to understand the impact of the disease on their mobility and to design appropriate rehabilitation interventions [32], [33].

Traditionally, gait analysis has relied on marker-based motion capture systems, which are considered the gold standard for accurate tracking of human motion [6]. However, these systems present limitations [3], [10]. Firstly, they require attaching multiple markers to the body, which can be time-consuming and inconvenient, especially for people with impairments. Moreover, the presence of markers may affect the naturalness of their movement [18], [21]. Additionally, marker-based systems are expensive and necessitate trained personnel to apply the markers correctly and process the data, making the analysis dependent on operators and resource-intensive. In fact, the accuracy of marker-based systems is highly dependent on the correct placement of markers, and any misplacement can lead to erroneous results. Furthermore, modifying the marker setup post-recording (*e.g.*, repositioning markers or adding new ones) is not feasible, limiting the flexibility of data collection and analysis.

Wearable sensors, *e.g.*, Inertial Measurement Units (IMUs), have been explored as a more affordable and non-invasive alternative and their potential has been demonstrated in analyzing gait in people with MS [23], [35]. However, wearable sensors still require direct contact with the body, which can limit comfort and introduce potential interference from objects [13]. Furthermore, choosing the optimal locations on the body to place the sensors is challenging [8], [24]. Video-based motion analysis offers a compelling alternative [4], [5], [11], [19], [30], eliminating the need for physical contact while enabling the acquisition of not only the motion itself but also the context in which the action occurs. These systems can also be fully automated, minimizing reliance on skilled operators and streamlining the data analysis process.

Human pose estimation algorithms, based on computer vision and deep neural networks, have shown significant progress in the last years [37]. These advancements have opened up possibilities for employing efficient methods to extract information from RGB video data for analyzing human motion, including gait analysis in clinical applications [4], [5], [11], [19], [30]. However, only in recent years quantitative studies have been performed to compare the information extracted from multi-views video-based markerless techniques with the gold standard marker-based systems, particularly concerning meaningful kinematic variables and spatio-temporal parameters of human gait [19], [30]. These studies presented the potential of markerless techniques with respect to gold standard motion capture system, highlighting the possibility to adopt these systems in clinical settings. Unfortunately, these methods have been assessed mainly with unimpaired participants, and, to the best of our knowledge, no attempts have been done to verify their potential for early detection of gait changes in individuals with MS. Therefore, in this manuscript, we leveraged a video-based approach [19] to extract quantitative information of the gait of 25 people with MS and 25 age and sex matched unimpaired controls. The algorithm utilizes RGB video data acquired from three viewpoints and calibration parameters to compute 3D keypoints positions. Then, this information is exploited to extract spatio-temporal and kinematic gait parameters. In a previous study, this pipeline was evaluated on unimpaired individuals and compared against a gold-standard marker-based system [19]. This previous work demonstrated that the three-camera markerless setup could reliably capture both joint kinematics and spatiotemporal parameters in unimpaired adults, with sufficient accuracy for clinical and research applications. Building upon these results, the present study investigate whether our video-based approach could distinguish between different populations in a real-world hospital environment, where gait was recorded during routine neurological visits without requiring additional preparation or specialized personnel for marker placement. Thus, we leveraged the above-mentioned markerless video-based gait analysis pipeline to examine gait patterns in individuals with multiple sclerosis (MS), focusing on two types of gait: normal walking and tandem gait. Tandem gait was included based on evidence from prior studies highlighting its utility in the early detection of balance impairments in individuals with MS [2], [17]. MS participants were divided into two groups based on their Expanded Disability Status Scale (EDSS) scores: a high EDSS group ($EDSS \geq 3$) and a low EDSS group ($EDSS < 3$). The primary aim of the study was to assess the effectiveness of video-based analysis in detecting gait deficits in MS during routine neurological visits, considering different EDSS scores.

II. Material and Methods

A. Participants

We prospectively collected gait data from 25 participants diagnosed with MS according to MC Donald criteria 2017 [31] (19 females, 6 males, mean age $\pm$ standard deviation: 40 $\pm$ 10, mean disease duration $\pm$ standard deviation: 10 $\pm$ 7.5 years, median EDSS [minimum, maximum]: 2 [1, 6]). Inclusion criteria included Expanded Disability Status Scale score (EDSS) ≤ 6 and adequate vision, with correction using glasses if necessary. Of the 25 participants with MS, 3 used an assistive device to walk during the experimental trials, as they regularly rely on such devices in their daily activities. To evaluate our pipeline's ability to detect different levels of impairments, we divided the dataset into two groups based on the EDSS score, following clinical guidelines and setting the threshold to 3. An EDSS score of ≥ 3 indicates moderate disability, while an EDSS < 3 corresponds to no or minimal disability. A similar approach was taken in [27], which set the threshold to 3.5. This choice allowed us to create two groups with a similar number of participants. The high EDSS group ($EDSS \geq 3$) was composed of 11 people (10 females, 1 males, mean age $\pm$ standard deviation: 48 $\pm$ 8, mean disease duration $\pm$ standard deviation: 14 $\pm$ 8 years, median Expanded Disability Status Scale score (EDSS) [minimum, maximum]: 5 [3, 6]), while the low EDSS group ($EDSS < 3$) was composed of 14 people (9 females, 5 males, mean age $\pm$ standard deviation: 34 $\pm$ 8, mean disease duration $\pm$ standard deviation: 7 $\pm$ 5 years, median Expanded Disability Status Scale score (EDSS) [minimum, maximum]: 2 [1, 2]). In the same context, we also acquired the gait of 25 unimpaired controls (age and sex matched with MS participants).

Prior to data collection, a power analysis was conducted to estimate the required sample size for the study. Preliminary gait data were collected from individuals in the three study groups: unimpaired controls, low-EDSS, and high-EDSS participants. For normal gait, stride time was selected as the primary parameter for estimating effect size. Based on preliminary data, an effect size of 0.71 was obtained. Using a significance level (α) of 0.05 and a desired power ($1 - \beta$) of 0.90, the analysis indicated a minimum total sample size of 30 participants. For tandem gait, heel-to-toe distance was identified as the primary outcome measure. An effect size of 1.55 was calculated from the preliminary dataset. With α set at 0.05 and power at 0.90, the estimated minimum sample size was 10 participants per group to detect significant differences. Power analyses were conducted using G*Power software. Based on these calculations, we enrolled 11 participants in the high-EDSS group, 14 in the low-EDSS group, and 25 age- and sex-matched unimpaired controls, ensuring adequate statistical power and reliability of the findings.

The study adhered to the principles of the Declaration of Helsinki and it was approved by the Institutional Ethical Committees (CER Liguria, approval No. 222REG2017 and the Review Board of the Department of Informatics, Bioengineering, Robotics, and Systems Engineering at the University of Genoa, Protocol No. CE DIBRIS-008/2020). Informed consent was obtained from all participants prior to their involvement in the study.

B. Experimental Setup and Protocol

The gait data were captured using a three-RGB-cameras setup, with the synchronized cameras (RGB Mako G125 GigE cameras with Sony ICX445 CCD sensor, temporal resolution: 30 frame per seconds (fps), spatial resolution: 1292 $\times$ 964) placed at three different viewpoints to allow for accurate 3D

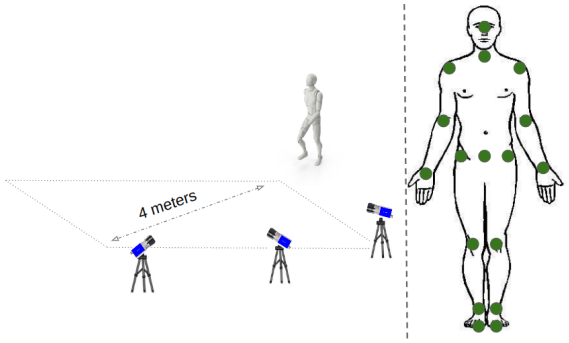

Fig. 1. Sketch of the setup adopted for data acquisition (left) and visual representation of the detected keypoints (in green) from the frontal viewpoint (right).

reconstruction of keypoints (see Figure 1 for a setup sketch). Each participant performed two types of gait: normal walking along a straight path, and tandem gait, which involves walking in a heel-to-toe manner along a straight line. For each gait modality, the participants walked in straight lines from one side of a room to the opposite and they were asked to walk as naturally as possible at their preferred speed. The video acquisition path was 4 m long and participants were required to perform 6 trials, 3 for each direction. The acquisition area did not capture at least one initial step before participants entered, nor one final step after they exited the acquisition path. To further minimize potential start-up and terminal effects, we excluded the first and last steps within the camera's field of view for each trial and analyzed three central gait cycles: two from the contralateral (non-initiating) leg and one from the initiating leg (*i.e.*, the second cycle). Consequently, at least two initial and two final steps were systematically excluded from the analysis to avoid introducing variability associated with gait initiation and termination [15]. This approach ensured that only gait cycles occurring in the middle portion of the walking task were considered and yielded a total of 18 gait cycles per participant.

C. Data Analysis Pipeline

Our analysis consists of the following key steps [19]: (a) keypoints detection using Pose ResNet-152, a state-of-the-art algorithm for pose estimation [34]; (b) refinement of the detections using Adafuse [36] and temporal filtering; (c) 3D reconstruction of keypoints positions based on a geometric approach [12]; (d) spatio-temporal and kinematic gait parameters computation using biomechanical modeling approaches (*e.g.*, Opensim [7]). In the following, we summarize the key aspects of each step. The overall accuracy of the spatio-temporal parameters and joint kinematics obtained from our markerless video-based method has been assessed in our previous validation study [19], where we compared these measures against a gold-standard motion capture system in unimpaired individuals.

1) Keypoints Detection: We used Pose ResNet-152 [34], a state-of-the-art convolutional neural network (CNN) designed for human pose estimation, as the backbone for detecting 2D keypoints in each video frame. This model was pre-trained on large-scale datasets and generates probability maps for each keypoint. Importantly, as highlighted in [19], this model do not consider keypoints on the feet, indispensable for our goal. For this reason, we first performed a fine tuning of the Pose ResNet-152 architecture considering a subset of the well-known Human3.6m dataset (we considered one image every 20 frames, for a total of 180,000 images) [14]. We did not perform any fine-tuning on our new MS dataset. Figure 1 shows a visual representation of the keypoints detected.

2) Keypoints Refinement and Filtering: To enhance the accuracy of the detected 2D keypoints, we applied the Adafuse algorithm [36], which leverages epipolar geometry to refine the keypoints detected by Pose ResNet-152. Adafuse integrates information from multiple camera viewpoints, improving keypoint detection by combining probability maps from adjacent views. This step is critical in minimizing errors, especially in occluded or difficult-to-detect body parts, by refining the 2D keypoint estimates. Then, we applied a low-pass Butterworth filter (4th order, 7 Hz cutoff frequency).

3) Keypoints 3D Reconstruction: Once the refined 2D keypoints were obtained, we reconstructed the 3D positions of these keypoints using a geometric approach. By triangulating the refined keypoints from multiple camera viewpoints and leveraging the intrinsic and extrinsic parameters obtained from camera calibration, we computed accurate 3D coordinates for each keypoint. This process allowed us to generate a full 3D skeleton of the body of each participant.

4) Spatio-Temporal and Kinematic Gait Parameters Computation: The 3D keypoints extracted from both people with MS and unimpaired controls were processed to quantify gait characteristics, including spatio-temporal and joint kinematic parameters. First, gait cycles were detected automatically for each participant. A gait cycle was defined as the period from one heel strike to the subsequent heel strike of the same foot. To detect the gait cycles, we analyzed the speed profiles of the heel keypoints, focusing on the vertical movement [25]. The gait cycles were defined as the intervals between two time instants where the heel's speed decreased below 10% of the local maximum value immediately following the peaks. Once the gait cycles were identified, we computed the spatio-temporal parameters for each participant. We computed the mean speed calculated as the average value over the gait cycle of the speed of the pelvic midpoint keypoint (see Figure 1). Then starting from the heels' trajectory, we calculated stride length, stride time, stance phase, swing phase, and step width. Stride length was measured as the distance traveled during a complete gait cycle. The stance phase was defined as the percentage of the gait cycle during which the foot was in contact with the ground, and the swing phase was its complement, representing the time when the foot was not in contact with the ground. Step width was calculated as the lateral distance between the midpoints of the left and right feet during a step. For tandem gait, during the double support phase (*i.e.*, when the velocity of both feet approached zero), the anteroposterior distance between the heel of the front foot and the toes of the back foot was computed to quantify heel-to-toe alignment. In addition to spatio-temporal parameters, joint angles were estimated using a biomechanical

TABLE I

NORMAL WALKING: SPATIO-TEMPORAL PARAMETERS COMPUTED FOR LOW-EDSS, HIGH-EDSS AND UNIMPAIRED GROUPS, AND STATISTICAL RESULTS OF THE ANOVA AND THE TUKEY POST-HOC TEST BETWEEN LOW-EDSS AND UNIMPAIRED (LAST ROW, FIRST VALUE FOR EACH COLUMN) AND BETWEEN HIGH-EDSS AND UNIMPAIRED (LAST ROW, SECOND VALUE FOR EACH COLUMN). WE REPORT THE MEAN ± THE STANDARD DEVIATION OF EACH PARAMETER. THE STANCE AND SWING PHASES ARE REPORTED IN % WITH RESPECT TO THE WHOLE GAIT CYCLE; STRIDE LENGTH AND STEP WIDTH AND EXPRESSED IN METERS (M); STRIDE TIME IN SECONDS (S); SPEED IN METERS PER SECONDS (M/S)

	Stance Phase (%)	Swing Phase (%)	Stride Length (m)	Step Width (m)	Stride Time (s)	Speed (m/s)
unimpaired	60.1 ± 3.3	39.9 ± 3.3	1.37 ± 0.25	0.10 ± 0.03	1.10 ± 0.04	1.25 ± 0.24
low EDSS	60.3 ± 2.9	39.7 ± 2.7	1.33 ± 0.30	0.12 ± 0.04	1.11 ± 0.05	1.20 ± 0.28
high EDSS	65.1 ± 3.5	34.9 ± 3.5	1.07 ± 0.38	0.15 ± 0.04	1.24 ± 0.07	0.86 ± 0.30
ANOVA	F(2,47)=9.29 p<0.001	F(2,47)=9.29 p<0.001	F(2,47)=3.95 p=0.026	F(2,47)=8.24 p<0.001	F(2,47)=28.62 p<0.001	F(2,47)=7.98 p=0.001
***p*-values Post-Hoc**	0.986/<0.001	0.986/<0.001	0.918/0.022	0.553/<0.001	0.845/<0.001	0.876/<0.001

model implemented in OpenSim [7]. OpenSim is widely employed in gait analysis as it facilitates the association of detected keypoints with biomechanical models of the human skeleton, enabling detailed kinematic and muscle activation analyses. In this study, we used the Rajagopal Model [28], a comprehensive full-body musculoskeletal model commonly applied in dynamic simulations of human movement. OpenSim offers two key tools tailored to this purpose: *Scaling* and *Inverse Kinematics*. The Scaling tool was applied to adapt a generic skeleton model to align with the input keypoints, while the Inverse Kinematics tool was employed to simulate skeleton motion and compute joint angles for each participant's gait cycle, providing insights into the dynamic movements. Particularly, we computed the hip flexion/extension, knee flexion/extension, ankle dorsi-/plantar-flexion, hip ab-/ad-duction, and pelvis tilt. Both the spatio-temporal parameters and the joint angles were averaged between left and right legs.

D. Statistical Analysis

As for normal gait, an analysis of variance (ANOVA) was conducted to assess differences in both spatio-temporal gait parameters and joint angle temporal profiles among the three study groups (unimpaired controls, low EDSS, and high EDSS), with group considered as a between-subjects factor. Following a significant main effect, we identified specific differences between group pairs (Tukey's post hoc test for the spatio-temporal parameters and pairwise t-test for the joint angles). Before performing the ANOVA, the normality assumption was verified with the Shapiro-Wilk test and the homogeneity assumption with the Levene's test. Tandem gait analysis was performed exclusively for the low-EDSS group, as it represents a more challenging task typically feasible only for individuals with lower levels of disability. In this case, an unpaired t-test was employed to assess statistically significant differences between low-EDSS and age- and sex-matched unimpaired participants, with the significance level set at $p < 0.05$. Statistical analysis were performed using the Jamovi environment (Jamovi software 2.6.44, jamovi.org) for spatio-temporal parameters. For the joint angles analysis, we used the Statistical Parametric Mapping (SPM) method, which is specifically designed to analyze time-varying signals. We applied this method to the 1-D spatio-temporal variables describing joint angles throughout the gait cycle, using the open-source software spm1d [26]. This approach allowed us to assess statistically significant differences in joint kinematic waveforms across the entire gait cycle while preserving the temporal structure of the data, rather than relying on discrete point comparisons. Statistical significance was set at $p < 0.05$.

III. RESULTS

A. Normal Gait

For normal gait, all spatio-temporal parameters showed significant differences across the high-EDSS, low-EDSS, and unimpaired control groups (see Table I). Also, the high-EDSS group exhibited altered gait patterns characterized by increased stride time, reduced stride length, slower gait speed, and a prolonged stance phase relative to the swing phase. However, no significant differences were observed between the low-EDSS group and the unimpaired controls, supporting the evidence that individuals with lower disability have more preserved gait function during normal walking.

The joint angle profiles were analyzed using a one-way ANOVA, which revealed significant effects (Figure 2 second column). Furthermore, groups comparison highlighted that the high-EDSS group showed significant differences from the unimpaired controls (Figure 2 fourth column). The high-EDSS group exhibited reduced flexion in the knee joints during the swing phase and reduced ankle dorsiflexion. Their pelvis was in anteroversion, reaching significantly higher values than controls at the beginning and at the end of the gait cycle. The high EDSS group also had a reduced range of motion (ROM) in the frontal plane: the abduction was lower between the 10% and the 30% of the gait cycle associated to a lower adduction between the 60% and the 65% of the gait cycle. In contrast, no significant differences were observed in the joint angles between the low-EDSS group and the unimpaired controls (Figure 2 third column).

B. Tandem Gait

In the tandem gait analysis, we focused on the low-EDSS group and compared their gait performance to the age and sex-matched unimpaired controls, as most individuals (8 out

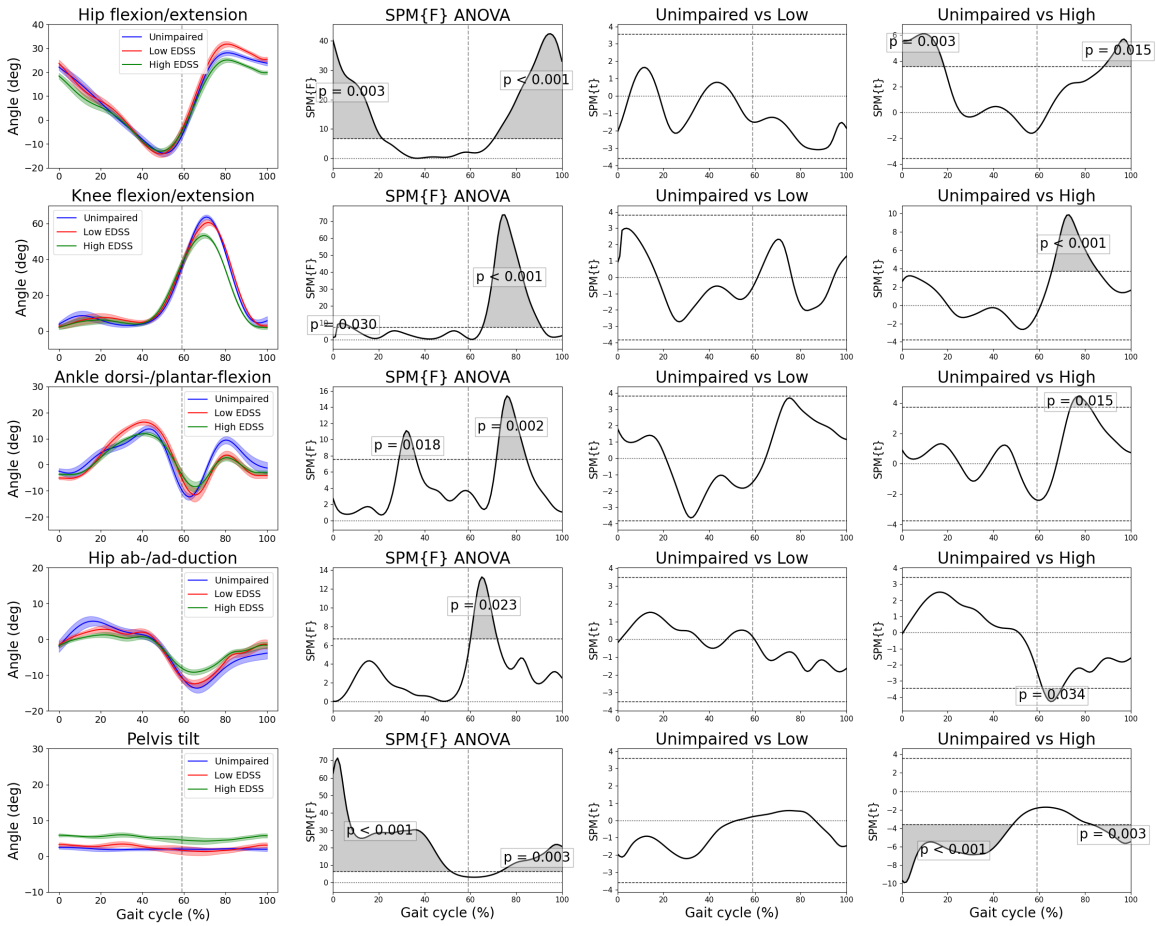

Fig. 2. First column: mean (solid lines) and standard error (shadow areas) of the joint angles for unimpaired (black), low-EDSS (red), and high-EDSS (green) participants. Second column: results of the ANOVA performed considering all the participants. Third column: group comparison between unimpaired and low-EDSS groups. Last column: group comparison between unimpaired and high-EDSS groups. From top to bottom: hip flexion/extension, knee flexion/extension, ankle dorsi-/plantar-flexion, hip ab-/ad-duction, and pelvis tilt.

TABLE II
TANDEM WALKING: STRIDE TIME (S) AND HEEL-TOE DISTANCE (M) COMPUTED FOR LOW-EDSS AND UNIMPAIRED GROUPS, AND STATISTICAL RESULTS (LAST ROW)

	Stride Time (s)	**Heel-to-Toe distance (m)**
unimpaired	1.97 ± 0.12	0.06 ± 0.04
low EDSS	2.08 ± 0.13	0.13 ± 0.06
t-test *p*-values	0.042	<0.001

of 11) with high EDSS scores were unable to perform tandem gait. By performing an unpaired t-test, we found significant differences in stride time between the low-EDSS group and unimpaired controls during tandem gait (Table II). Furthermore, people with MS in the low-EDSS group consistently exhibited a larger space between the heel and the toe, indicating less precise foot placement, a large base of support and reduced ability to maintain the heel-to-toe position.

IV. DISCUSSION AND CONCLUSION

This study highlights the potential of video-based markerless systems for detecting and quantifying gait impairment in people with MS. Our findings show that this approach can successfully identify significant gait abnormalities, particularly in individuals with higher disability scores (high-EDSS group). Additionally, tandem gait analysis revealed subtle gait deficits even in the low-EDSS group, underscoring its sensitivity in detecting early-stage impairments and distinguishing between different levels of impairment, as supported by previous studies [2], [17]. The fact that these differences were also observable using a video-based gait analysis approach is particularly encouraging, as it highlights the potential of markerless video analysis to detect clinically relevant gait alterations in a non-cumbersome and accessible manner.

Our analysis of normal gait revealed significant differences in both joint kinematics and spatio-temporal parameters between the high-EDSS group and unimpaired controls. The high-EDSS group exhibited reduced joint mobility in the sagittal plane, particularly in the ankle in the swing/pre-swing phase and knee during the swing phase, along with an increased anteroversion of the pelvis and reduced ROM of the hip in the frontal plane. These results are consistent with previous studies showing that individuals with greater MS-related disability tend to experience a slower, more cautious gait, characterized by shorter steps and a prolonged stance phase to compensate for balance and coordination issues [29]. In contrast, no significant differences were observed between the

low-EDSS group and the unimpaired controls during normal gait. Aside from potential errors associated with the markerless method (discussed below), this finding may suggest that individuals with lower levels of disability retain relatively normal gait patterns under standard walking conditions. On one side, this finding is in line with existing literature [29], which has shown that gait abnormalities may not be easily detectable in early-stage MS during straightforward walking tasks. However, on the other side, there are studies that highlight that wearable inertial sensors [22] and marker-based motion capture systems [1] detected early gait alterations in minimally impaired MS individuals, even when global clinical scales like the EDSS show no significant changes. Wearable sensors provide detailed, continuous data on gait, which allows for the detection of subtle deviations that might be overlooked by traditional methods or video analysis. These sensors are especially useful in capturing dynamic changes in gait across different conditions, such as fatigue, offering a more thorough understanding of gait abnormalities in MS. Marker-based motion capture systems offer higher spatial and temporal resolution than markerless approaches, allowing for the detection of subtle motor impairments that may go unnoticed with less precise techniques. This reinforces the importance of validating markerless systems against reference-standard technologies to assess their sensitivity and potential clinical utility, especially when investigating subtle motor changes in clinical populations.

In contrast to normal gait, tandem gait analysis was able to reveal subtle yet significant differences between the low-EDSS group and unimpaired controls. Tandem gait is a more challenging task that requires greater balance and coordination, and it is particularly useful for detecting early deficits in motor control. In our study, low-EDSS individuals had a larger space between the heel and toe than unimpaired controls. This increased distance indicates difficulties in maintaining precise foot placement.

The extension of this approach to more prolonged walking assessments, such as the 6-minute walk test, deserves further investigation. Applying the method in such contexts would require careful evaluation of the camera's spatial resolution over extended distances, as well as consideration of the increased setup complexity. Incorporating additional cameras may help maintain tracking accuracy over a larger area, but could also reduce the system's portability and introduce greater intrusiveness. Wearable sensors remain more suitable for outdoor settings or for monitoring walking over extended distances, where deploying and calibrating a large number of cameras would be impractical.

Although our results are promising, several limitations should be acknowledged. First, the sample size of the MS cohort was relatively small, particularly for the tandem gait analysis. This may limit the generalizability of our findings and warrants further validation in larger and more diverse MS populations. Future studies should include broader cohorts to confirm our results and better assess their applicability across varying clinical profiles. Second, while previous work has highlighted that toe foot keypoint estimated via markerless pose estimation algorithms are among the most susceptible to noise [20], it is important to contextualize this limitation. Although motion blur can arise during rapid movements, the average gait speed and the typical frequency of human locomotion are well below half the 30 Hz frame rate of our video recordings. Given that participants walked at a natural pace, motion blur is not expected to have introduced significant errors in our analysis. Indeed, as discussed in [20], any impact of motion blur was minor and mostly confined to the feet toe, without meaningfully affecting the heel keypoint used for gait cycle detection. Moreover, our previous studies compared gait event detection using the same video-based pipeline against gold-standard systems such as OptiTrack [19] and SMART DX (with integrated force platforms) [20], and found strong consistency in key spatiotemporal parameters, such as stride time and stride length. These findings support the robustness of our video-based methodology in capturing reliable gait metrics. Additionally, while our study focused on gait differences between people with MS and unimpaired controls, future research could explore how these gait impairments evolve over time or in response to rehabilitation interventions.

Also accounting for this limitation the ability to detect gait abnormalities using a non-cumbersome, markerless video-based system holds significant potential for clinical practice. Traditional marker-based systems are often expensive, invasive, and require specialized equipment and expertise, limiting their use in routine clinical settings. In contrast, video-based systems offer a cost-effective, user-friendly alternative that can be easily integrated into clinical workflows. Moreover, these systems can capture gait in more natural and unconstrained environments, which may lead to a more meaningful assessment of mobility in people with MS. Our findings suggest that video-based gait analysis can be particularly useful for monitoring disease progression in MS. In people with higher disability scores, the algorithm can clearly detect significant gait abnormalities, while in people with lower EDSS scores, tandem gait analysis may reveal early deficits in balance and coordination that would otherwise go unnoticed.

Institutional Review Board Statement

The study adhered to the principles of the Declaration of Helsinki and it was approved by the Institutional Ethical Committees (CER Liguria, approval No. 222REG2017 and the Review Board of the Department of Informatics, Bioengineering, Robotics, and Systems Engineering at the University of Genoa, Protocol No. CE DIBRIS-008/2020). Informed consent was obtained from all participants prior to their involvement in the study.

References

[1] M. G. Benedetti, R. Piperno, L. Simoncini, P. Bonato, A. Tonini, and S. Giannini, "Gait abnormalities in minimally impaired multiple sclerosis patients," *Multiple Sclerosis*, vol. 5, no. 5, pp. 363–368, May 1999.

[2] I. Carpinella et al., "Balance impairments in people with early-stage multiple sclerosis: Boosting the integration of instrumented assessment in clinical practice," *Sensors*, vol. 22, no. 23, p. 9558, Dec. 2022.

[3] B. Carse, B. Meadows, R. Bowers, and P. Rowe, "Affordable clinical gait analysis: An assessment of the marker tracking accuracy of a new low-cost optical 3D motion analysis system," *Physiotherapy*, vol. 99, no. 4, pp. 347–351, Dec. 2013.

[4] A. Castelli, G. Paolini, A. Cereatti, and U. D. Croce, "A 2D markerless gait analysis methodology: Validation on healthy subjects," *Comput. Math. Methods Med.*, vol. 2015, no. 1, Jul. 2015, Art. no. 186780.

[5] A. Cimorelli, A. Patel, T. Karakostas, and R. J. Cotton, "Validation of portable in-clinic video-based gait analysis for prosthesis users," *Sci. Rep.*, vol. 14, no. 1, p. 3840, Feb. 2024.

[6] S. L. Colyer, M. Evans, D. P. Cosker, and A. I. T. Salo, "A review of the evolution of vision-based motion analysis and the integration of advanced computer vision methods towards developing a markerless system," *Sports Med.-Open*, vol. 4, no. 1, pp. 1–15, Dec. 2018.

[7] S. L. Delp et al., "OpenSim: Open-source software to create and analyze dynamic simulations of movement," *IEEE Trans. Biomed. Eng.*, vol. 54, no. 11, pp. 1940–1950, Nov. 2007.

[8] S. Díaz, J. B. Stephenson, and M. A. Labrador, "Use of wearable sensor technology in gait, balance, and range of motion analysis," *Appl. Sci.*, vol. 10, no. 1, p. 234, Dec. 2019.

[9] N. E. Fritz, R. E. R. Marasigan, P. A. Calabresi, S. D. Newsome, and K. M. Zackowski, "The impact of dynamic balance measures on walking performance in multiple sclerosis," *Neurorehabilitation Neural Repair*, vol. 29, no. 1, pp. 62–69, Jan. 2015.

[10] L. Garello et al., "A study of at-term and preterm infants' motion based on markerless video analysis," in *Proc. 29th Eur. Signal Process. Conf. (EUSIPCO)*, Chiara Tacchino, France, Aug. 2021, pp. 1196–1200.

[11] X. Gu, F. Deligianni, B. Lo, W. Chen, and G. Z. Yang, "Markerless gait analysis based on a single RGB camera," in *Proc. IEEE 15th Int. Conf. Wearable Implant. Body Sensor Netw. (BSN)*, Mar. 2018, pp. 42–45.

[12] R. Hartley and A. Zisserman, *Multiple View Geometry Comput. Vis.* Cambridge, U.K.: Cambridge Univ. Press, 2003.

[13] K. Homan, K. Yamamoto, K. Kadoya, N. Ishida, and N. Iwasaki, "Comprehensive validation of a wearable foot sensor system for estimating spatiotemporal gait parameters by simultaneous three-dimensional optical motion analysis," *BMC Sports Sci., Med. Rehabil.*, vol. 14, no. 1, p. 71, Dec. 2022.

[14] C. Ionescu, D. Papava, V. Olaru, and C. Sminchisescu, "Human3.6M: Large scale datasets and predictive methods for 3D human sensing in natural environments," *IEEE Trans. Pattern Anal. Mach. Intell.*, vol. 36, no. 7, pp. 1325–1339, Jul. 2014.

[15] Y. Jian, D. Winter, M. Ishac, and L. Gilchrist, "Trajectory of the body COG and COP during initiation and termination of gait," *Gait Posture*, vol. 1, no. 1, pp. 9–22, Mar. 1993.

[16] F. D. Lublin et al., "Defining the clinical course of multiple sclerosis: The 2013 revisions," *Neurol.*, vol. 83, no. 3, pp. 278–286, 2014.

[17] C. Massot et al., "Upper limb contribution during tandem gait in multiple sclerosis: An early marker of balance impairments," *J. Biomechanics*, vol. 149, Mar. 2023, Art. no. 111492.

[18] M. Moro, M. Casadio, L. A. Mrotek, R. Ranganathan, R. Scheidt, and F. Odone, "On the precision of markerless 3D semantic features: An experimental study on violin playing," in *Proc. IEEE Int. Conf. Image Process. (ICIP)*, Sep. 2021, pp. 2733–2737.

[19] M. Moro, G. Marchesi, F. Hesse, F. Odone, and M. Casadio, "Markerless vs. marker-based gait analysis: A proof of concept study," *Sensors*, vol. 22, no. 5, p. 2011, Mar. 2022.

[20] M. Moro, G. Marchesi, F. Odone, and M. Casadio, "Markerless gait analysis in stroke survivors based on computer vision and deep learning: A pilot study," in *Proc. ACM Symp. Appl. Comput.*, Jul. 2020, pp. 2097–2104.

[21] M. Moro et al., "A markerless pipeline to analyze spontaneous movements of preterm infants," *Comput. Methods Programs Biomed.*, vol. 226, Nov. 2022, Art. no. 107119.

[22] R. Müller, D. Hamacher, S. Hansen, P. Oschmann, and P. M. Keune, "Wearable inertial sensors are highly sensitive in the detection of gait disturbances and fatigue at early stages of multiple sclerosis," *BMC Neurol.*, vol. 21, no. 1, pp. 1–8, Sep. 2021.

[23] R. Müller, D. Hamacher, P. M. Keune, and P. Oschmann, "Stability of sensor-based gait parameters reassessed after a period of one year in people with multiple sclerosis," *BMC Neurol.*, vol. 23, no. 1, p. 120, Mar. 2023.

[24] R. Müller, L. Schreff, L.-E. Koch, P. Oschmann, and D. Hamacher, "Measuring gait stability in people with multiple sclerosis using different sensor locations and time scales," *Sensors*, vol. 21, no. 12, p. 4001, Jun. 2021.

[25] C. M. O'Connor, S. K. Thorpe, M. J. O'Malley, and C. L. Vaughan, "Automatic detection of gait events using kinematic data," *Gait Posture*, vol. 25, no. 3, pp. 469–474, Mar. 2007.

[26] T. C. Pataky, J. Vanrenterghem, and M. A. Robinson, "Zero- vs. one-dimensional, parametric vs. non-parametric, and confidence interval vs. hypothesis testing procedures in one-dimensional biomechanical trajectory analysis," *J. Biomechanics*, vol. 48, no. 7, pp. 1277–1285, May 2015.

[27] M. Pau et al., "Inter-joint coordination during gait in people with multiple sclerosis: A focus on the effect of disability," *Multiple Sclerosis Rel. Disorders*, vol. 60, Apr. 2022, Art. no. 103741.

[28] A. Rajagopal, C. L. Dembia, M. S. DeMers, D. D. Delp, J. L. Hicks, and S. L. Delp, "Full-body musculoskeletal model for muscle-driven simulation of human gait," *IEEE Trans. Biomed. Eng.*, vol. 63, no. 10, pp. 2068–2079, Oct. 2016.

[29] G. Severini et al., "Evaluation of clinical gait analysis parameters in patients affected by multiple sclerosis: Analysis of kinematics," *Clin. Biomechanics*, vol. 45, pp. 1–8, Jun. 2017.

[30] K. Song, T. J. Hullfish, R. Scattone Silva, K. G. Silbernagel, and J. R. Baxter, "Markerless motion capture estimates of lower extremity kinematics and kinetics are comparable to marker-based across 8 movements," *J. Biomechanics*, vol. 157, Aug. 2023, Art. no. 111751.

[31] A. J. Thompson et al., "Diagnosis of multiple sclerosis: 2017 revisions of the McDonald criteria," *Lancet Neurol.*, vol. 17, no. 2, pp. 162–173, Dec. 2017.

[32] K. L. Williams and S. G. Brauer, "Walking impairment in patients with multiple sclerosis: The impact of complex motor and non-motor symptoms across the disability spectrum," *Austral. J. Gen. Pract.*, vol. 51, no. 4, pp. 215–219, Apr. 2022.

[33] T. A. L. Wren, C. A. Tucker, S. A. Rethlefsen, G. E. Gorton, and S. Õunpuu, "Clinical efficacy of instrumented gait analysis: Systematic review 2020 update," *Gait Posture*, vol. 80, pp. 274–279, Jul. 2020.

[34] B. Xiao, H. Wu, and Y. Wei, "Simple baselines for human pose estimation and tracking," in *Proc. Eur. Conf. Comput. Vis. (ECCV)*, 2018, pp. 466–481.

[35] A. Zahn et al., "Validity of an inertial sensor-based system for the assessment of spatio-temporal parameters in people with multiple sclerosis," *Frontiers Neurol.*, vol. 14, Apr. 2023, Art. no. 1164001.

[36] Z. Zhang, C. Wang, W. Qiu, W. Qin, and W. Zeng, "AdaFuse: Adaptive multiview fusion for accurate human pose estimation in the wild," *Int. J. Comput. Vis.*, vol. 129, no. 3, pp. 703–718, Mar. 2021.

[37] C. Zheng et al., "Deep learning-based human pose estimation: A survey," 2020, *arXiv:2012.13392*.