# Cardiac Autonomic Activity in Commercial Aircrew During an Actual Flight Duty Period

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BACKGROUND:

The work schedules of airline crewmembers include extended workdays, compressed work periods, and limited time for recovery, which may lead to cardiovascular strain and fatigue. The aim of this study was to evaluate changes in heart rate variability (HRV) during work and sleep, and with respect to work characteristics and breaks.

**METHODS:** 

We followed 49 airline crewmembers during four consecutive workdays of ≥39 h. Data included HRV measurements, a questionnaire, and sleep/work diaries. HRV parameters include root mean square of successive differences (RMSSD), standard deviation of the normal beat-to-beat differences (SDNN), and the low and high frequency ratio (LF/HF).

RESULTS:

The results indicate higher levels of cardiovascular strain on the 4th compared to the 1st workday, most prominent among cabin crewmembers. In this group, we observed indications of decreased cardiovascular strain by increasing duration of sleep, demonstrated by increased RMSSD (B=2.7,95% CI 1.6, 3.8) and SDNN (B=4.4,95% CI 3.0, 5.7), and decreased LF/HF (B=-0.2,95% CI, -0.4,-0.01). Similarly, longer duration of breaks was associated with lower cardiovascular strain, indicated by increased RMSSD (B=0.1,95% CI 0.03, 0.1) and SDNN (B=0.1,95% CI 0.1, 0.1). Among pilots, increased LF/HF indicated higher cardiovascular strain in those who often or always reported of high workload (B=4.3,95% CI 2.3, 6.3; and B=7.3,95% CI 3.2, 11.4, respectively).

DISCUSSION:

The results support the contention that the studied work period increases cardiac strain among airline crew. Work characteristics, breaks, and sleep are associated with changes in HRV.

**KEYWORDS:** 

airline crew, extended working hours, fatigue, heart rate variability.

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s shown in a review by Keclund and Axelsson, <sup>16</sup> a considerable number of studies have documented negative impact of nonstandard working time arrangements regarding sleep, fatigue, cardiovascular health, performance, and safety. Schedules of airline pilots and cabin crew are extremely irregular, and involve early starts, long daily working hours, compressed working weeks, short rest periods, all of which may contribute to sleep disorders, strain, and fatigue.<sup>2</sup> Disruption of sleep alters sleep-wake timing and destabilizes physiology.<sup>35</sup>

The autonomic nervous system (ANS) is the primary regulator of heart rate, and there is growing evidence for its role in development of a wide range of diseases.<sup>31</sup> The ANS consists of two major branches: the sympathetic, associated with energy mobilization, and the parasympathetic, associated with vegetative and restorative functions. With long-term strain and incomplete recovery, protracted activation of the sympathetic

part of the ANS may potentially increase the risk of cardiovascular diseases (CVD).<sup>17</sup> The sympathetic and parasympathetic branches act antagonistically to preserve the dynamic equilibrium of the vital functions, but may become unbalanced in response to external or internal stimuli or demands. In the cardiovascular system, this dynamic regulation results in the variation of the time intervals between consecutive heart beats, so called heart rate variability (HRV). HRV reflects the balance of the cardiovascular system; sympathetic activity tends to

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increase heart rate (HR) and to decrease HRV, whereas parasympathetic activity decreases HR and increases HRV.<sup>31</sup>

Measuring HRV is a noninvasive procedure. The HRV measurement is considered a reliable estimator of the ANS status, which enables indirect observation of subtle changes due to stress, strain and recovery.<sup>31</sup> A normal subject shows a good degree of variation of the heart rate, reflecting a good capability to react to external stimuli.<sup>5</sup>

Various occupational factors are believed to modulate workers' cardiovascular health.<sup>32</sup> However, it is unclear to which degree the observed association between irregular working hours and increased risk of CVD is a result of psychological and physiological strain related to the long working hours or other work environment factors, or to unhealthy lifestyle as a result of shift work, (smoking, poor diet quality, and lack of physical activity) which could influence CVD.<sup>33</sup> For studies of shift work and health, it has been recommended to include, in addition to objective assessment of working time, variables concerning sleep,<sup>16</sup> physiological mechanisms, and perceived work-stress.<sup>18</sup> Furthermore, recovery may be evaluated by the variations in HRV during sleep after work and leisure time.

The aim of this study was to evaluate changes in HRV during an actual flight duty period and sleep, and with respect to work characteristics and breaks.

# **METHODS**

# **Subjects**

The subjects of this study were pilots and cabin crewmembers in a commercial airline based in Norway. Employees in the airline were informed of the study and encouraged to participate in an e-mail from the airline's management of flight and cabin operations and the worker's unions. Subsequently, representatives from the study's project group were present at the crew base at Oslo airport on several occasions to recruit crewmembers. Initially 160 crewmembers agreed to participate. The main criteria for selection of the final sample were characteristics of the planned flight duty period (FDP) of the enrolled crew-

members. Every month, personal schedules for the coming 4 wk are presented to all crewmembers. Schedules eligible for the present study were those including a 4-d work period of at least 39 work hours, in which the first workday was at least 10 h, and including only short-haul flights operated by Boeing 737 aircraft. Work periods consisting of 4 d of flight were chosen, as these were the most common among cabin crewmembers at the time we started our data collection. Most pilots with a variable roster pattern would also have a majority of 4-d

work periods. The > 39 h for the 4-d period was chosen as this represented a compressed work period. The > 10 h first workday was chosen to focus on workdays well exceeding the 7.5–8 h limits for normal workdays in the country where these airline crewmembers are based. We chose short-haul operations only, as we sought to avoid night work and time differences. We sought to ensure as similar work procedures as possible, thus the study was limited to only one aircraft type.

The final sample consisted of 59 healthy airline crewmembers, 18 pilots (16 men and 2 women) and 41 cabin crewmembers (6 men and 35 women). The subject characteristics are shown in **Table I**. A cross-shift/cross-week design, in which the subjects served as their own controls, was applied. The study period was a work period of 4 d. The study protocol was approved by the Regional Committee for Medical Research Ethics (2014/1508/REK sør-øst B), and the subjects provided a written informed consent.

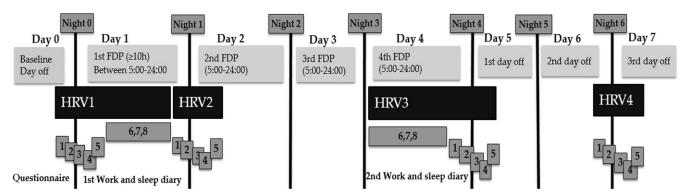
# Procedure

A flowchart of the data collection is shown in Fig. 1. At baseline, the evening before the actual work period, the subjects completed a questionnaire concerning age, weight, height, health status, physical activity, work experience, and work characteristics. The questionnaire included questions from validated questionnaires such as QPS-Nordic<sup>22</sup> and Bergen Insomnia Scale.<sup>23</sup> During the first and the fourth workday, the subjects completed a work and sleep diary, including check-in and check-out times for duty, commuting time, time and duration of breaks, and irregularities of flights (such as delays). Furthermore, the diary included questions about the duration and quality of sleep. The quality of sleep was determined by a simplified version of the Bergen Insomnia Scale, including reporting episodes of over 30 min before falling asleep, over 30-min awake periods during the night, and early awakening more than 30 min earlier than desired. The Samn-Perelli Fatigue Score (SP)<sup>26</sup> was included in the diary, and the subjects stated their alertness at check-in time, after 8 h, and at check-out time, according to the following scores: 1) Fully alert, wide awake; 2) Lively, responsive, not at peak; 3) OK, somewhat fresh;

**Table I.** Subject Characteristics (N = 59).

	<b>PILOTS (</b> <i>N</i> = 17*)	CABIN CREWMEMBERS (N = 41)
Men	15	6
Women	2	35
Current smoker	1	1
Age	52 (SD 12.3)	40 (SD 7.4)
Body Mass Index (kg/m²)	25 (SD 2.8)	24 (SD 3.6)
Reported sleep disturbances last year	9 (50%)	21 (51%)
Reported sleep disturbances last month	5 (27%)	12 (12%)
Work experience (in years)	26 (SD 7.7)	17 (SD 11.4)
Commuting time – one way (in minutes)	70	57
Physical activity (1–3 h per week)	11 (65%)	24 (59%)
Physical activity (> 3 h per week)	6 (35%)	17 (41%)
Reported work/family conflict	7 (39%)	18 (44%)
Content with work pattern	8 (44%)	27 (66%)
Reported high workload quite often/always	11 (62%)	12 (31%)
Control over important decisions never/seldom	8 (44%)	22 (56%)

 $<sup>^{\</sup>ast}$  Questionnaires were returned by 17 of 18 pilots.



**Fig. 1.** Flowchart of the data collection.

4) A little tired, less than fresh; 5) Moderately tired, let down; 6) Very tired, difficulty concentrate; 7) Completely exhausted.

HRV was measured by eMotion 3D-sensors (Mega Electronics Ltd, in Kuopio, Finland) to assess cardiovascular strain during flight duty, and recovery during leisure time and sleep. The sensor contained an accelerometer, which was utilized to detect onset of sleep, and time of awakening. At baseline, the evening before the first day of flight duty, a member of the research group demonstrated the application and activation of the HRV-sensors, and also activated the first sensor (HRV1) (Fig. 1). The sensor was deactivated, and replaced by a second sensor (HRV2) at check-out time after the first day of flight duty. HRV2 was deactivated by the subject before his/her flight duty on the second work day. On the morning of the fourth workday, the subject activated a third sensor (HRV3), and deactivated it the next morning. The subjects were instructed to activate a fourth sensor (HRV4) at bedtime the evening of the second day off, and deactivate it the next morning. HRV measurements were preprocessed and analyzed using the Kubios HRV analysis software.<sup>30</sup> The selected HRV measures were chosen according to the guidelines of the Task Force of the European Society of Cardiology<sup>12</sup> and are described in Table II. HRV data were visually inspected, to exclude artifacts such as missing beats or ectopic beats. Subsequently, 5-min time segments free of artifacts were selected from each work hour during the day, from the first 4 h of sleep, and from the wake-up hour in the morning. Finally, hourly mean HRV values were calculated from the selected 5-min time segments. In Fig. 1, boxes numbered 1, 2, 3, 4 represent the mean of 5-min time segments during the first 4 h of sleep, box 5 shows the time of awakening, and box 6, 7, 8 represent the working hours during the days.

The following comparisons were made for each HRV-parameter: 1) the difference between the first and fourth workday; 2) the difference between the baseline night (night 0) and the nights 1, 4 and 6; and 3) the differences between cabin crewmembers and pilots. Comparisons of 1), 2) and 3), with data stratified by work characteristics, showed duration of breaks, reported decision latitude, and perceived workload.

# **Statistical Analyses**

We applied linear mixed models to each of the HRV measurements for the hours of the workday, the hours of sleep, and for the time of awakening. A random intercept was included for

each subject. On the basis of existing knowledge of factors associated with cardiovascular disease, <sup>19,29</sup> we adjusted for the following variables: sex, age, and body mass index (BMI). We performed additional analyses adjusted for varying HR by adding a linear and quadratic term. In the present study, the work hours relate to the starting time of the workday, and not the clock hours, as check-in and check-out times vary considerably between the subjects. In mixed model analyses, we chose to collapse work hours 1–3, 4–7, and the work hours 8 until the end of the workday, in order to focus on main trends, and to improve statistical power (few subjects ended their workday at very late hours).

In separate analyses, we studied the effect of workplace characteristics, sleep duration, and duration of breaks during the workdays. Fisher's exact test was used to evaluate potential differences between flight commanders and first officers regarding job control and cardiovascular strain. Linear mixed models were analyzed using Stata 15. SPSS' Statistical Package for Windows 24.0 (SPSS Inc, Chicago, IL, USA) was applied for the univariate analyses of the self-reported data from the questionnaire and diaries.

**Table II.** Description of the Selected Heart Rate Variability Measurements.

HRV PARAMETER (UNIT)	DESCRIPTION AND INTERPRETATION
Mean RR interval (ms)	Mean of selected beat to beat (RR) interval series inversely proportional to HR.
RMSSD (ms)	Square root of the mean squared differences between successive RR intervals. RMSSD evaluates differences between successive RR intervals and reflects short-term variations. Low value indicates high cardiovascular strain.
HF powers (ms <sup>2</sup> )	High-frequency power (range 0.15–0.4 Hz). High HF indicates low cardiovascular strain.
SDNN (ms)	Standard deviation of normal heartbeat intervals. Estimates overall HRV, not distinguishing between changes due to reduced vagal tone or increased sympathetic activity. Low value indicates high cardiovascular strain.
LF power (ms <sup>2</sup> )	Low frequency power (range 0.04–0.15 Hz). High LF may indicate high cardiovascular strain.
LF/HF ratio	The selected frequency-domain parameter is the ratio between low frequency and high frequency power components. The LF/HF ratio estimates sympatho- vagal balance. High LF/HF indicates high cardiovascular strain

# **RESULTS**

The median number of years the participating pilots have worked in the airline is 26 yr (SD 7.7), and the cabin crewmembers 17 yr (SD 11.4). The distribution of socio-demographic variables are shown in Table I. Reported information from work and sleep diaries are shown in **Table III**.

The majority of the subjects in both professional groups reported that they felt quite refreshed at the start of both the first and the fourth day of the work period (not shown). The mean SP fatigue score was 2.2 both days, where 2 represents "lively, responsive, not at peak." However, the mean score at check-out time, at both the first and fourth workday, indicated tiredness; SP = 4.2 and 4.4 for the cabin crew, and 4.6 and 4.2 for the pilots, where score 4 represents "a little tired, less than fresh" and 5 "moderately tired, let down."

Irregularities and change of scheduled flights for the subjects resulted in work periods not meeting the criteria for inclusion, and thus reduced the number of subjects. While HRV1 was measured by 40 cabin crewmembers and 16 pilots, HRV2 was made by 38 cabin crewmembers and 18 pilots. For HRV3, the number of subjects was reduced to 24 cabin crewmembers and 12 pilots. HRV4 was measured by 19 cabin crewmembers and 12 pilots. The reduction in the sample during the work period was due to severe delays, diversions and rescheduling of flights, and to sick leave. In addition, some measurements, particularly day-time measurements, were excluded due to poor quality of the data.

Changes of HRV variables on workday 4 vs. workday 1 are shown in **Table IV**. **Fig. 2** shows the changes in HRV among the cabin crewmembers.

Mean RR was significantly increased during the first 7 h of workday 4 among cabin crewmembers, while among the pilots, we observed a significant decrease in mean RR during the first 3 h and after 7 h of duty on workday 4. RMSSD showed a nonsignificant decrease during all working hours of workday 4 among the pilots, and after the third work hour among cabin crewmembers, when compared to workday 1.

SDNN decreased significantly among the cabin crewmembers after the third hour of duty on workday 4 compared to workday 1, while only a nonsignificant decrease was observed among the pilots. A significantly higher LF/HF was seen in the cabin crewmembers on the after the third work hour throughout workday 4. The only observed significant differences

between pilots and cabin crewmembers were lower RR among the pilots during the first 3 h (B = -78.7, 95% CI -112.1, -45.3), and during the next 4 h of workday 4 (B = -39.2, 95% CI -69.0, -9.4).

**Table V** shows changes of HRV variables during the nights after the first and the fourth workday, and after two days off, compared with the baseline night (night 0). Changes are tabulated for each of the first 4 h of sleep, and for the hour of awakening.

Among cabin crewmembers, we observed a significant increase of mean RR at the time of awakening both in the morning after night 1, after night 4, and after night 6, when compared to awakening after night 0 (before first workday). Mean RR among the pilots reveals a similar pattern as that for cabin crewmembers, with a significant increase at the time of awakening both in the morning after workday 1 and 4, and after 2 d off. For RMSSD among cabin crewmembers, a significant increase was only observed at the time of awakening after night 6, compared to night 0. Among the pilots, RMSSD was significantly increased at the third hour of sleep in night 6. For SDNN among both cabin crewmembers and pilots, there was no difference for the awakening hours after any of the later nights, when compared with the awakening time after night 0. Neither did we observe any significant change of SDNN during sleep in any of the nights 1, 4, and 6, compared to night 0. LF/HF was significantly lower among cabin crewmembers at the time of awakening after nights 1, 4, and 6, when compared to awakening after night 0, while no difference was observed during sleep. Among the pilots, LF/HF decreased significantly during the second and third hour of sleep the night after workday 1 (night 1), compared to the night before workday 1 (night 0). A significant decrease of LF/HF was also seen during the second hour of sleep in the night after the 4-d work period (night 4), and during the second and third hour of sleep in the night after 2 d off (night 6). Observed differences between pilots and cabin crewmembers during the nights comprise a lower LF/HF among the pilots from the fourth hour of sleep (B = -1.6, 95% CI -3.1, -0.2) until awakening (B = -2.1, 95% CI -3.8, -0.5) the night after workday 1. The pilots also showed a lower LF/HF than the cabin crewmembers from the second, third, and fourth hour of sleep the night after the last workday; (B = -2.5, 95% CI - 4.0, -1.1) (B = -2.0, 95% CI - 3.7, -0.3) (B = -2.2, 95% CI -4.1, -0.3), respectively.

Among cabin crewmembers, during all of the work shifts, RMSSD, SDNN, and LF/HF were all significantly associated

**Table III.** Reported Information from the Work/Sleep Diaries.

	CABIN CREWMEMBERS		PILOTS		
	Day 1	Day 4	Day 1	Day 4	
	(N = 41)	(N = 26)	(N = 18)	(N = 16)	
Mean hours of sleep the previous night (range, hours)	6 (4–8)	7 (4–8)	7 (4.5–8)	7 (4.5–10)	
No. of subjects reporting ≥ 30 min before falling asleep	16 (39%)	4 (10%)	5 (28%)	1 (6%)	
No. of subjects reporting ≥ 30 min awake in between sleep	13 (32%)	7 (17%)	3 (17%)	3 (17%)	
No. of subjects reporting awakening ≥ 30 min before planned	16 (39%)	14 (34%)	5 (28%)	4 (22%)	
Mean check-in times AM (SD)	7:45(2.0)	9:19 (2.6)	8:13 (2.5)	10:2 (3.3)	
Mean duty hours (SD)	11.2 (1.1)	10.6 (1.8)	10.9 (1.3)	10.1 (1.6)	
Mean no. of flight sectors (range)	3.8 (2-6)	3.5 (2-5)	3.4 (2-6)	3.9 (3-5)	

Table IV. Changes in Mean RR, RMSSD, SDNN and LF/HF Between Workday 4 and 1, for Different Time Segments, Among Cabin Crewmembers and Pilots.

	CABIN CREWMEMBERS (N = 35)				PILOTS (N = 12)			
	MEAN RR	RMSSD	SDNN	LFHF	MEAN RR	RMSSD	SDNN	LFHF
Hours	B (95% CI)	B (95% CI)	B (95% CI)	B (95% CI)	B (95% CI)	B (95% CI)	B (95% CI)	B (95% CI)
0-3	29.4 (13.8,45.0)	3.3 (-0.2, 6.7)	-1.0 (-5.1, 3.2)	0.0 (-0.5, 0.5)	-48.7 (-82.9, -14.5)	-2.7 (-7.4, 1.9)	-3.2 (-7.5, 1.1)	-0.2 (-1.0, 0.6)
4-7	14.3 (0.2, 28.3)	-1.6 (-4.8, 1.5)	-4.9 (-8.7,-1.1)	0.5 (0.1, 1.0)	-24.4 (-54.8, 6.1)	-0.6 (-4.7, 3.6)	-1.4 (-5.2, 2.4)	-0.2 (-1.0, 0.5)
8+	-11.3(-26.2, 3.7)	-3.2 (-6.5, 0.1)	-5.2 (-9.2,-1.2)	0.6 (0.1, 1.1)	-37.0 (-72.4, -1.7)	-1.6 (-6.4, 3.2)	-2.4 (-6.8, 2.1)	0.3 (-0.5, 1.2)

B (95% CI): estimate of difference between Day 4 and Day 1 (Day 4 – Day 1) with 95% confidence interval, adjusted for gender, age and BMI. Bold numbers indicate significant values.

with the duration of sleep the previous night, when adjusted for sex, age, and BMI. An increase of RMSSD (B = 2.7, 95% CI 1.6, 3.8) and SDNN (B = 4.4, 95% CI 3.0, 5.7), and a decrease of LF/HF (B = -0.2, 95% CI, -0.4, -0.01) were observed by increasing number of sleeping hours. Among the pilots, we did not observe similar significant changes for any of these parameters. Among the cabin crewmembers, we also observed an increase in mean RR (B = 0.2, 95% CI 0.6, 0.3), RMSSD (B = 0.1, 95% CI 0.03, 0.1) and SDNN (B = 0.1, 95% CI 0.1, 0.1) by increasing duration of breaks, when adjusted for sex, age, and BMI. However, among the pilots, no increase was observed for any of these parameters.

Reported demand/control factors, such as workload and decision latitude, were associated with variations in LF/HF among the pilots. LF/HF was significantly lower (B = -4.9, 95% CI -8.7, -1.2) in pilots who rarely perceived the workload as heavy, while an increased LF/HF was seen in pilots who

often (B = 4.3, 95% CI 2.3, 6.3) or always (B = 7.3, 95% CI 3.2, 11.4) perceived the workload as heavy. A somewhat different pattern was observed regarding decision latitude. LF/HF was decreased both workday 1 and 4 among pilots who reported of rarely being able to influence decisions important for their work (B = -3.5, 95% CI -6.6, -0.3). A decrease was also observed in pilots who reported of often being able to influence decisions important for their work (B = -5.9, 95% CI -10.0, -1.7). Among cabin crewmembers, the demand/control issues were not associated with any significant changes of the HRV parameters.

# **DISCUSSION**

In the present study, we identified HRV measures indicating a higher level of cardiovascular strain on the fourth, compared to

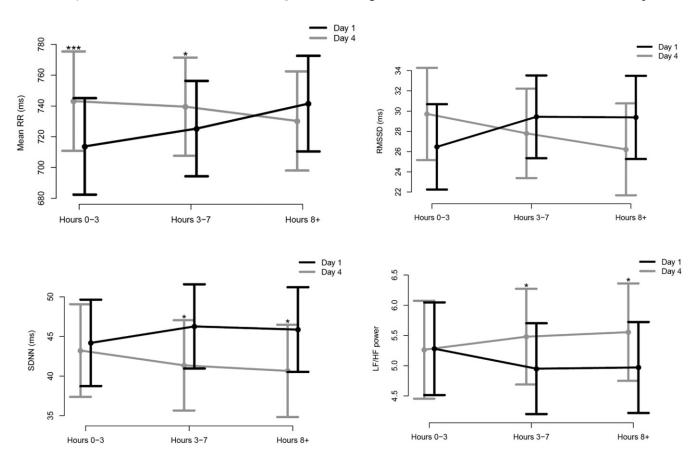


Fig. 2. RR, RMSSD, SDNN, LF/HF on workday 4 vs. workday 1 among cabin crewmembers.

**Table V.** Changes in Mean RR, RMSSD, SDNN and LF/HF Between the Baseline Night (night 0) and the Nights 1, 4 and 6, Respectively, for Each of the First Four Hours of Sleep and the Hour of Wake-Up, Among Cabin Crewmembers and Pilots.

		CAB	IN CREWMEMI	BERS (N = 41)		PILOTS ( <i>N</i> = 17)			
NIGHT	HOURS	Mean RR B (95% CI)	RMSSD B (95% CI)	SDNN B (95% CI)	LFHF B (95% CI)	Mean RR B (95% CI)	RMSSD B (95% CI)	SDNN B (95% CI)	LFHF B (95% CI)
1 vs. 0	1 h sleep	0.1 (-32.9, 33.1)	1.6 (-4.3, 7.4)	0.6 (-4.2, 5.4)	-0.1 (-0.8, 0.7)	-30.4 (-85.6, 24.9)	-0.7 (-7.8, 6.4)	-0.5 (-8.5, 7.5)	-1.0 (-2.4, 0.3)
1 vs. 0	2 h sleep	-16.2 (-48.9, 16.6)	0.0 (-5.9, 5.8)	-1.1 (-5.9, 3.7)	-0.3 (-1.1, 0.4)	-9.0 (-64.1, 46.1)	1.8 (-5.3, 8.9)	-0.5 (-8.5, 7.4)	-1.9 (-3.2, -0.6)
1 vs. 0	3 h sleep	-12.3 (-45.0, 20.5)	-2.9 (-8.7, 2.9)	-4.6 (-9.4, 0.2)	0.1 (-0.7, 0.8)	-12.4 (-69.4, 44.6)	-0.8 (-8.1, 6.6)	-4.2 (-12.5, 4.0)	-2.5 (-3.8, -1.1)
1 vs. 0	4 h sleep	-12.3 (-45.3, 20.7)	-3.3 (-9.2, 2.5)	-3.0 (-7.8, 1.8)	0.2 (-0.5, 1.0)	-20.9 (-79.1, 37.4)	-4.1 (-11.6, 3.4)	-5.6 (-14.0, 2.9)	-0.7 (-2.1, 0.7)
1 vs. 0	wake-up	143.3 (91.9, 194.8)	4.0 (-5.2, 13.1)	-2.1 (-9.6, 5.4)	-1.5 (-2.7, -0.3)	118.6 (20.8, 216.3)	6.3 (-6.3, 18.9)	9.4 (-4.8, 23.5)	-2.0 (-4.3, 0.4)
4 vs. 0	1 h sleep	17.6 (-20.9, 56.0)	3.9 (-2.9, 10.8)	2.8 (-2.8, 8.4)	-0.2 (-1.1, 0.6)	4.0 (-56.7, 64.7)	3.9 (-4.0, 11.7)	4.2 (-4.6, 13.0)	-0.9 (-2.4, 0.6)
4 vs. 0	2 h sleep	-14.3 (-52.7, 24.2)	0.3 (-6.5, 7.2)	0.0 (-5.6, 5.6)	0.3 (-0.6, 1.2)	25.0 (-35.0, 84.9)	6.4 (-1.3, 14.2)	3.0 (-5.7, 11.7)	-1.9 (-3.3, -0.4)
4 vs. 0	3 h sleep	-10.4 (-48.8, 28.0)	2.8 (-4.0, 9.6)	2.2 (-3.4, 7.8)	0.4 (-0.5, 1.3)	9.8 (-53.6, 73.2)	5.4 (-2.8, 13.6)	4.9 (-4.3, 14.0)	-1.6 (-3.1, 0.0)
4 vs. 0	4 hsleep	-0.8 (-39.3, 37.6)	1.9 (-4.9, 8.8)	1.5 (-4.1, 7.1)	0.0 (-0.9, 0.9)	-21.5 (-84.6, 41.6)	-1.5 (-9.7, 6.6)	-3.1 (-12.2, 6.0)	-0.8 (-2.4, 0.7)
4 vs. 0	wake-up	185.8 (124.4, 247.2)	6.7 (-4.2, 17.6)	-0.6 (-9.6, 8.3)	-2.1 (-3.5, -0.7)	212.6 (115.3, 310.0)	3.9 (-8.7, 16.4)	4.8 (-9.3, 18.9)	-1.6 (-3.9, 0.8)
6 vs. 0	1h sleep	14.9 (-26.6, 56.5)	0.1 (-7.3, 7.5)	-1.9 (-8.0, 4.2)	-0.1 (-1.1, 0.8)	29.0 (-37.9, 95.9)	1.2 (-7.4, 9.8)	1.1 (-8.6, 10.8)	-1.1 (-2.8, 0.5)
6 vs. 0	2 h sleep	-18.4 (-60.9, 24.1)	-4.5 (-12.0, 3.0)	-4.9 (-11.1, 1.2)	-0.3 (-1.3, 0.7)	57.5 (-8.3123.3)	4.7 (-3.7, 13.2)	3.0 (-6.5, 12.5)	-1.7 (-3.3, -0.1)
6 vs. 0	3 h sleep	-12.7 (-55.6, 30.3)	-1.0 (-8.7, 6.6)	-0.8 (-7.0, 5.5)	0.3 (-0.7, 1.3)	44.0 (-29.5117.5)	12.2 (2.7, 21.6)	8.5 (-2.1, 19.1)	-1.9 (-3.7, -0.1)
6 vs. 0	4 h sleep	6.2 (-36.8, 49.2)	-1.8 (-9.4, 5.9)	-2.7 (-8.9, 3.6)	0.0 (-1.0, 1.0)	1.0 (-67.7, 69.8)	-0.1 (-9.0, 8.7)	-1.1 (-11.1, 8.8)	0.2 (-1.4, 1.9)
6 vs. 0	wake-up	244.5 (179.0, 309.9)	16.9 (5.2, 28.5)	8.1 (-1.5, 17.6)	-3.1 (-4.6, -1.6)	282.2 (174.1, 390.3)	4.5 (-9.4, 18.5)	6.3 (-9.4, 21.9)	-1.1 (-3.7, 1.5)

B (95% CI): estimate of difference between Night 1, 4, 6 and Night 0 (Night 1 – Night 0, Night 4-Night 0, Night 6-Night 0) with 95% confidence interval adjusted for gender, age, and BMI. Bold numbers indicate significant values.

the first workday, most prominent among the cabin crewmembers. This became apparent both through decreased SDNN, and an increased LF/HF after the first 3 h of duty on the fourth compared to the first workday. RR was increased during the first hours of the flight duty on the fourth compared to the first workday, which could indicate that the cabin crewmembers were rested when starting their duty on workday 4. This is in line with their subjective reports of feeling alert and is further supported by their reporting of less sleep disturbances and longer sleep duration the night before workday 4. Anticipation of high demands, or having to rise early, may have disturbed the sleep the night before the first day of flight duty. The night before workday 4, 50% of the cabin crewmembers stayed overnight in hotels, allowing them to recover undisturbed. Instead of the usual long commuting time, a hotel-to-airport transportation of short duration was at hand. This, and the anticipation of the days off ahead, may partly explain the lower report of sleep disturbances and increased duration of sleep the last night of the duty period.

Cardiovascular strain seemed to decrease with longer duration of sleep before the workdays, and with increasing number and duration of breaks among the cabin crewmembers. We did not observe any correlation between reported work characteristics and HRV-parameters among cabin crewmembers. A higher overall bodily stress, as measured by HRV parameters, was observed on the fourth vs. the first workday, in spite of the shorter mean duty length, and fewer flight sectors on the fourth day. This may have been influenced by the suboptimal environment in which cabin crewmembers perform their duty, handling heavy service trolleys in tiny galleys and narrow aisles, sometimes even uphill. Being in the service frontline, and attending to several hundred passengers during each day of flight duty, represents a psychological strain.<sup>6,34</sup> The continuous exposure to high levels of noise in the aircraft may further cause a shift in cardiovascular regulation toward sympathetic

dominance.<sup>8</sup> Days of duty in short-haul operations usually include multiple flight sectors, with a high number of passengers, repeated safety and service procedures, and represent a high workload. The accumulation of work hours, the combined physical and mental workload during the work period, may have contributed to the increased cardiovascular strain the fourth workday among the cabin crewmembers, in line with earlier research.<sup>13</sup>

For pilots, we observed a somewhat different trend. A decreased mean RR was found on workday 4 compared to workday 1, particularly during the morning and evening hours. In combination with the decreasing trends of RMSSD and SDNN all through the fourth day, this either indicates lack of recovery, or accumulation of strain from workday 1 to workday 4. The pilots reported of later check-in time for duty, less sleep disturbance and the same sleep duration the night preceding the fourth workday, when most of them stayed overnight in a hotel. However, self-reported recovery is not necessarily reflected in physiological recovery as indicated by HRV, as stressors that may lead to HRV-changes indicating increased cardiovascular strain may not result in similar subjective experience of stress.<sup>13</sup>

The lower RR among the pilots compared to cabin crewmembers during the first 7 h of duty on day 4 compared to day 1 was the only significant differences between the two groups of subjects, in spite of their very different work content.

We attempted to disentangle the effect of HR on HRV by adjusting for HR in the analyses of RMSSD, SDNN, and LF/HF. The differences between the first and the fourth workday were reduced but remained statistically significant for SDNN and LF/HF among the cabin crewmembers.

Previous research has shown a correlation between self-reported psychological strain and physiological indicators of strain as measured by HRV.<sup>15</sup> Similarly, in the present study, the reports of a high workload were often or always associated with

increased LF/HF. High job demands, in combination with low work control, is a potential determinant of reduced HRV, and the effect of high job strain has been associated with reduced HRV.

A person's ability to influence what happens in their work environment is a key element in handling work related strain. In the current study, associations were found between very low and very high decision latitude and decreased cardiovascular strain. The participating pilots were either commanders or first officers. While the commanders have full control of decisions, including the ultimate responsibility during a flight, the first officers have less control and less responsibility. A Fischer's exact test of any potential effect of the job category did not, however, reveal any difference between the two groups in how they reported on the question regarding decision latitude.

Operating an aircraft requires substantial cognitive effort and attention from the pilot, and the take-offs and landings represent the highest cognitive demand and workload. A Changes of HRV are related to both information processing and performance, and appear to be sensitive to increased risk of mental overload. The decreased RR during the first and the last hours of workday 4 may partly be explained by take-offs and landings during these hours. A study of the same subjects revealed that the number of take-offs and landings was associated with increased reaction times as measured by neurobehavioral tests, indicating increased fatigue, thus supporting that take-offs and landing represent high workload.

The HRV measurements during nights among the subjects in both professional groups indicate a satisfactory recovery after work. Slow wave sleep, mainly occurring during the first 4 h of sleep, is related to recovery<sup>4</sup> and we, therefore, analyzed HRV parameters from each of these hours. We observed greater sympathetic activation of the ANS during work than during sleep, in line with earlier research. <sup>10,14</sup> This may be due in part to the strong influence of physical activity on the circadian changes in HR.<sup>3</sup> HRV is also influenced by posture, as HRV recorded in the standing position shows higher LF/HF values compared to the supine position. <sup>20</sup>

In a study of pathways from circadian strain to morbidity, Puttonen et al.<sup>25</sup> concludes that shift work can induce psychological circadian strain, due to a disrupted work/life balance. While 39% of the cabin crewmembers in the present study and 44% of the pilots reported a work/family conflict, no significant associations were observed between such conflict and any of the HRV parameters, neither among cabin crewmembers nor among pilots (not shown).

Both cabin crewmembers' and pilots' ability to achieve sufficient sleep, in spite of the irregular working hours, may probably be considered a healthy worker effect.<sup>27</sup> While the included subjects in the course of their long work experience may have developed appropriate ways to handle challenges in their work plans regarding sleep and recovery, airline crewmembers suffering from severe sleep problems due to the irregular work hours have probably quit this type of work.

In the final analyses, we adjusted for sex, age, and BMI. Generally, while HRV is observed to be lower in women under age

30 compared to men, sex differences gradually disappear between age 30 and age 50. In the present study however, sex did not seem to influence any of the HRV parameters. With regard to age, older subjects had an overall lower HRV, in line with earlier research.<sup>36</sup> The quite high mean age of the subjects in the present study reflects the age distribution among flight personnel in the company, which is partly a result of several periods of hiring freeze after the turn of the century. The observed decreased HRV by increasing BMI is in accordance with results from previous studies.<sup>19</sup> Previous studies have shown that smoking disrupts the normal ANS functioning, characterized by increased sympathetic drive and reduced HRV, and parasympathetic modulation. There were two smokers among the subjects. We performed analyses with and without the smokers, and as only minor differences between smokers and nonsmokers were observed, we decided to keep the smokers in the final analyzed dataset. In the present study, the impact of physical activity could not be evaluated, as all subjects reported a medium to high level of physical activity.

The strengths of the present study include the use of a crossover design that eliminates uncontrolled confounding by use of an external control group.<sup>28</sup> Furthermore, the study was conducted in a real-life situation in which we had detailed exposure information, and information of other factors related to work and leisure time, which may potentially influence HRV parameters.<sup>32</sup> One limitation of the study was the small sample size, particularly the small pilot group, which reduced statistical power and thus the capacity to detect differences and trends observed at a borderline statistical significance. This was partly modified by the repeated-measurement design. Furthermore, the study population was not a random sample, which may have resulted in selection bias, and decreased the generalizability of the results. The skewed gender distribution within the two professions is however, similar to the actual distribution within the group of cabin crewmembers and pilots in most airlines. Finally, although though the number of work hours was similar among all subjects, the exact times for checkin and check-out for duty varied, which may also have influenced the results.

The findings of this study supports the contention that a work period consisting of a minimum of 39 working hours during 4 d increases cardiac strain among cabin crewmembers and pilots. Higher cardiovascular strain was observed on the fourth vs. the first day of flight duty, most prominent in the cabin crewmembers. Analyses of HRV during the nights indicate a satisfactory recovery after the first and the fourth workday in both professional groups. Among the pilots, high demands were associated with increased cardiovascular strain during the entire work period. Among the cabin crewmembers, increased duration of sleep before, and breaks during the workdays, reduced cardiovascular strain. Further research is required to disentangle the complex interplay between predictors of cardiovascular health, such as work hours, work content, breaks, sleep, and factors related to the organizational work environment.

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