CLINICAL STUDY

The acute effects of cigarette smoking on human optic nerve head and posterior fundus circulation in light smokers

YASUHIRO TAMAKI, MAKOTO ARAIE, MIYUKI NAGAHARA, KEN TOMITA, MASAO MATSUBARA

Abstract

Purpose To study the acute effects of cigarette smoking on tissue circulation in the optic nerve head (ONH) and posterior fundus in smokers with a short smoking history.
Methods Ten healthy smokers whose length of smoking history was within 2 years (age 25 ± 1 years; smoking index (number of cigarettes smoked per day × length of smoking history in years) 16 ± 4, mean ± SE) were included in the study. Using the laser speckle method, normalised blur (NB) value, a quantitative index of tissue blood velocity, was measured every 0.125 s and averaged over three pulses across an area located in the temporal site of the ONH free of visible surface vessels (NB_{ONH}) and across an area located halfway between the macula and the ONH with no discrete vessels visible ($NB_{ch\text{-}ret}$). NB_{ONH}, $NB_{ch\text{-}ret}$ and intraocular pressure (IOP) in one randomly chosen eye, and blood pressure (BP) and pulse rate (PR) were measured before, and 1, 5, 10, 15, 20, 25, 30, 45, 60 and 90 min after sham smoking. One week later, NB_{ONH}, $NB_{ch\text{-}ret}$ and IOP in the same eye, and BP and PR were measured after cigarette smoking according to the same time schedule as in the control experiment.
Results After sham smoking, no parameter showed a significant change during the experiment. Differences in NB_{ONH} from the baseline were not significant between the smoking experiment and sham smoking experiment, while $NB_{ch\text{-}ret}$ showed a significant difference at 30 min. Inter-group difference in the time course of the difference from baseline was significant (ANOVA, $p = 0.0246$, 0.0021). BP and PR were significantly increased between 1 and 20 min after smoking while IOP showed no significant change at any time of measurement.
Conclusions In light smokers, cigarette smoking showed little effect on tissue blood velocity in the ONH and slightly decreased that in the posterior fundus, suggesting a significant increase in vascular resistance in these tissues.

Key words Cigarette smoking, Fundus circulation, Laser speckle method, Light smoker, Optic nerve head

Previous studies have suggested that cigarette smoking, which is a risk factor for atherosclerotic complications in the coronary, aortic and cerebral circulation,[1] causes various circulatory changes in several organs through the local metabolic and vascular effects of systemically absorbed products.[2-5] Thus, the effect of cigarette smoking on the ocular circulation is of clinical interest.

There have been several previous reports studying the effect of cigarette smoking on the ocular circulation in habitual smokers. Using the blue field simulation technique, Robinson *et al.*[6] reported that macular leucocyte velocity in habitual smokers was increased by about 12% immediately after smoking. Using the laser speckle method, we have recently found that smoking increases tissue blood velocity in the optic nerve head (ONH) and possibly that in the choroid, in habitual smokers whose length of smoking history was 10 years on average.[7] On the other hand, Morgado *et al.*,[8] using laser Doppler velocimetry and retinal photography, reported that smoking caused a 10% decrease in retinal blood flow rate in habitual smokers. The former two techniques measure leucocyte velocity in capillaries in the macular region and blood velocity in the peripheral circulation, respectively, as opposed to volumetric blood flow through large retinal vessels. In rabbits, a single inhalation of cigarette smoke[9] or a single subcutaneous injection of 2.5 mg/kg nicotine[10] significantly reduced choroidal blood flow by about 20%, while it showed no significant change after long-term (16 or 25 weeks) inhalation of mainstream smoke[11] or 12 or 20 weeks twice-daily subcutaneous injection of

Y. Tamaki
M. Matsubara
Eye Clinic
Tokyo Metropolitan Geriatric Hospital
Tokyo, Japan

M. Araie
M. Nagahara
Department of Ophthalmology
University of Tokyo School of Medicine
Tokyo, Japan

K. Tomita
Eye Clinic
Tokyo Kosei Nenkin Hospital
Tokyo, Japan

Makoto Araie, MD ✉
Department of Ophthalmology
University of Tokyo School of Medicine
7-3-1, Hongo
Bunkyo-ku
Tokyo 113, Japan

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2.5 mg/kg nicotine.[12] Further, there is some evidence that the change in vascular tone with cigarette smoking may be different in non-smokers from that in chronic smokers,[13] and it has been shown that during test smoking, forearm vascular resistance increases significantly in healthy non-smokers[14] whereas it remains unchanged in healthy, chronic smokers.[15] Thus, the effect of smoking on the ocular circulation might be different according to the duration of the habit. There have been no previous reports, however, on the effect of smoking on the ocular circulation exclusively in smokers with a short smoking history.

In the present study we investigated the effects of cigarette smoking on the ONH and posterior fundus circulation in smokers with a short smoking history for 90 min after smoking, using the non-invasive laser speckle method.

Materials and methods

Instrument and measurement in human ONH or posterior fundus

A fundus camera was equipped with a diode laser (wavelength 808 nm) and image sensor (100 × 100 pixels). The laser beam was focused on the fundus, which was illuminated by a halogen lamp. The scattered laser light was imaged on an image sensor corresponding to an area of 1.06 × 1.06 mm (when imaging with 45° visual angle of the fundus camera) in the human fundus, on which a speckle pattern appeared. The difference between the average of the speckle intensity (I_{mean}) and the speckle intensity for successive scannings of the image speckles at the pixels on the sensor plane was calculated, and the ratio of I_{mean} to this difference was defined as normalised blur (NB).[16] NB is nearly equivalent to the reciprocal of speckle contrast defined by Fercher and Briers[17,18] and a quantitative index of tissue blood velocity. The NB was calculated by the logic board every 0.125 s successively for 7 s and divided into 50 colour-coded levels, which were displayed as colour graphics on a colour monitor showing the two-dimensional variation in NB level over the measurement field.[19] The average NB level (NB_{av}) in any rectangular field of interest can be calculated and the change in NB_{av} over 7 s can be monitored and stored on a MO disc at one time.

Thirty minutes before the measurements, the pupil was dilated with one drop of Mydrin M (0.4% tropicamide, Santen Pharmaceutical, Osaka, Japan). During the measurement the subject was asked to watch a target light; an electrocardiogram was monitored simultaneously. The image speckles from the measurement field located in the temporal site of the ONH, corresponding to a field of 0.72 × 0.72 mm (30° visual angle of the fundus camera), were recorded when fixation was satisfactory and two-dimensional variation of the NB level over the measurement field (colour map) was stored on a MO disc. Using the data stored on the MO disc, the average NB across the largest rectangular field in the measurement field free of visible surface vessels, further averaged over three cardiac pulses, was calculated to obtain NB_{ONH} (Fig. 1). The size of this rectangular field varied among subjects to avoid surface vessels, ranging from 0.22 × 0.32 mm (30 × 45 pixels) to 0.36 × 0.50 mm (50 × 70 pixels). Image speckles from a field located halfway between the macula and the ONH with no discrete retinal vessels visible, corresponding to a field of 1.06 × 1.06 mm (45 ° visual angle), were recorded and average NB across the largest rectangular field free of visible surface retinal vessels was averaged over three cardiac pulses to obtain $NB_{ch\text{-}ret}$ in the same manner as above. The size of this rectangular field varied among subjects to avoid surface visible retinal vessels, ranging from 0.48 × 0.64 mm (45 × 60 pixels) to 0.64 × 0.74 mm (60 × 70 pixels). The movement of the subject's eye in any direction during the measurement period was checked by the method previously described.[20] It was also checked by inspecting the colour map taken every 0.125 s; when there was no eye movement during measurement, visible surface vessels did not change position on the colour map.[19] On the other hand, when eye movement occurred, visible surface vessels in the colour map changed position according to the eye movement.

Subjects

A total of 10 young volunteers who had no systemic or ocular disease and only mild refractive errors were included in the study. Informed written consent was obtained after the nature of the study had been explained fully. Ages of the subjects ranged from 21 to 31 years, averaging 25 years. The number of cigarettes smoked per day averaged 11 ± 3, and the length of the subjects' smoking history averaged 1 ± 0.2 year (mean ± SE). The smoking index (number of cigarettes smoked per day × length of smoking history in years) was 16 ± 4. Subjects were instructed strictly to refrain from smoking and drinking beverages containing caffeine for at least 8 h prior to each experiment. No serum tests were performed to confirm abstinence from these two habits.

Sham smoking and actual smoking experiments

In the control experiment, after dilating the pupil the image speckles from the ONH, those from the posterior fundus and intraocular pressure (IOP) in one randomly chosen eye, brachial arterial blood pressure and pulse rate (PR) were recorded at 1100 hours. Mean brachial arterial blood pressure (BP_m) was calculated by:

$$BP_m = BP_d + 1/3(BP_s - BP_d) \qquad (1)$$

where BP_d and BP_s are diastolic and systolic brachial arterial blood pressure, respectively. Thereafter, sham smoking was performed using a short drinking straw as a cigarette substitute for 5 min at 1120 hours. The image speckles from the ONH and those from the posterior fundus recordings in the same measurement site were carried out between 1 and 2, 5, 10, 15, 20, 25, 30, 45, 60 and 90 min after sham smoking and the IOP measurements 10, 30, 60 and 90 min after sham smoking. After the experiment, a colour polaroid photograph was taken to record the site of measurement. One week later, the image speckles from the ONH and those from the posterior fundus in the same site of the same eye were recorded. IOP, BP_m and PR measurements were performed before and after smoking one standard brand cigarette (nicotine, 1.3 mg; tar, 15 mg) at the same time of day and according to the same time schedule as above. The subjects were required to smoke one entire cigarette within a 5 min period.

Two-dimensional variation of NB level over the measurement field (colour map) obtained from the subject's eye were all stored on MO discs as described above. A masked investigator determined the NB_{ONH} and $NB_{ch\text{-}ret}$ from the colour maps, referring to the colour polaroid photograph recording the site of measurement. In so doing, images of visible surface retinal vessels which were colour-coded as red on the colour map were used as markers to determine the site from which NB_{ONH} or $NB_{ch\text{-}ret}$ was to be obtained.

Data analysis

The time course of the difference in NB_{ONH} and $NB_{ch\text{-}ret}$ before and after sham smoking or actual smoking (ΔNB_{ONH}, $\Delta NB_{ch\text{-}ret}$), and the difference in IOP, BP_m and PR (ΔIOP, ΔBP_m and ΔPR) were plotted. Analysis of variance (ANOVA) for the data obtained by sequential measurements was used to detect inter-group difference. For exploratory purposes, the values obtained at each time point were compared with Student's *t*-test.

Results

In the control experiment, NB_{ONH}, $NB_{ch,\text{-}ret}$, BP_m, PR and IOP averaged 22.5 ± 1.0, 25.1 ± 1.2, 80.0 ± 2.8 mmHg, 68 ± 3/min and 11.7 ± 0.7 mmHg (mean ± SE) before sham smoking, respectively, and showed no significant change during the experimental period (Figs. 1, 2). In the actual smoking experiment, the same data averaged 21.8 ± 1.9, 25.7 ± 1.1, 80.9 ± 2.2 mmHg, 70 ± 2/min, and 11.5 ± 0.7 mmHg, respectively, and showed no significant inter-group difference from the data obtained in the control experiment ($p > 0.40$).

The NB_{ONH} after cigarette smoking tended to decrease, but no significant inter-group difference was seen at any time point ($p > 0.05$). ANOVA showed that the time course of NB_{ONH} was significantly different between after smoking and after sham smoking ($p = 0.024$ (group), $p < 0.001$ (group × time) (Fig. 1).

Inter-group difference in $\Delta NB_{ch\text{-}ret}$ was significant at 30 min after smoking ($p = 0.008$). ANOVA showed that the time course of $\Delta NB_{ch\text{-}ret}$ was significantly different between after smoking and after sham smoking ($p = 0.002$ (group), $p < 0.001$ (group × time)) (Fig. 1).

Compared with the value measured in the control experiment, both BP_m and PR significantly increased between 1 and 20 min after smoking compared with control ($p = 0.001$–0.041, Fig. 2). IOP showed no significant change (Fig. 2). Time courses of ΔBP_m and ΔPR were significantly different between after smoking and sham smoking ($p < 0.001$ (group), $p < 0.001$ (group × time), ANOVA, respectively).

Discussion

Method of measurement

According to Koelle *et al.*,[21] the penetration depth of near-infrared laser (wavelength 811 nm) in the cat optic nerve exceeds 1 mm. Thus, using the present apparatus, the effective depth of sampling in the ONH tissue will be greater than 1 mm. Using the same type of apparatus, NB_{av} obtained from the ONH and the ONH tissue blood flow rate determined with the hydrogen gas clearance method (where a hydrogen electrode was inserted into the ONH tissue to a depth of approximately 0.7 mm)

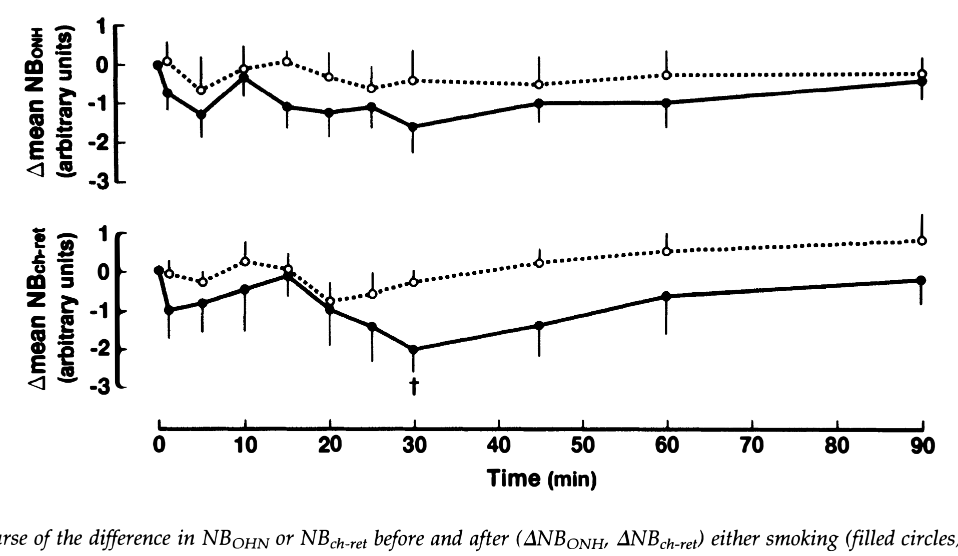

Fig. 1. *Time course of the difference in NB_{OHN} or $NB_{ch\text{-}ret}$ before and after (ΔNB_{ONH}, $\Delta NB_{ch\text{-}ret}$) either smoking (filled circles) or sham smoking (open circles). Each plot represents the mean value of 10 subjects. Error bar represents SEM. $NB_{ch\text{-}ret}$ was significantly decreased at 30 min after smoking (p = 0.008, paired* t*-test). †p < 0.01 by* t*-test for paired data for the difference from control (sham smoking experiment).*

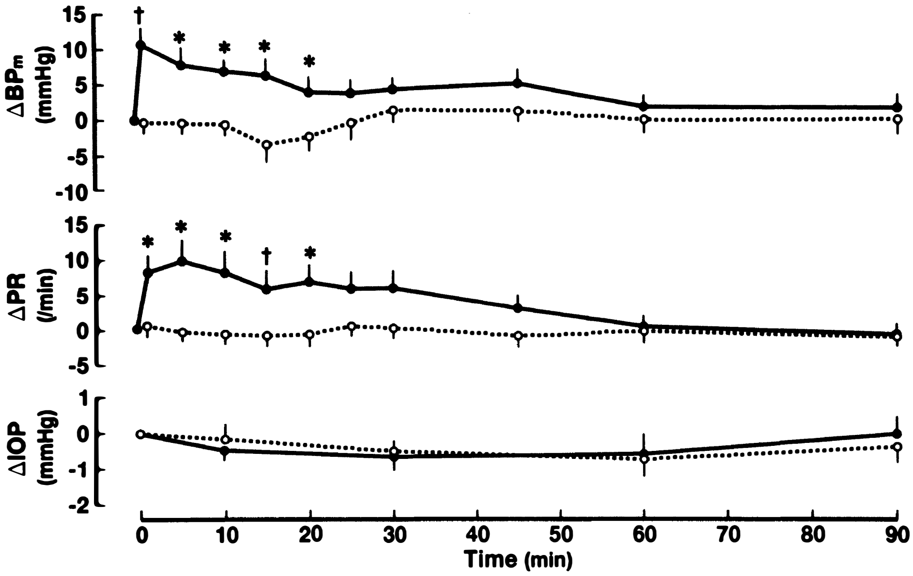

Fig. 2. *Time course of the difference in mean blood pressure (BP_m), pulse rate (PR) and intraocular pressure (IOP) before and after (ΔBP_m, ΔPR, ΔIOP) either smoking (filled circles) or sham smoking (open circles). Each plot represents the mean value of 10 subjects. Error bar represents SEM. BP_m and PR were significantly increased between 1 and 20 min after smoking as compared with control* ($p < 0.041$–0.001). *$p < 0.5$, †$p < 0.01$ *by* t*-test for paired data for the difference from control (sham smoking experiment).*

were compared in rabbit eyes under various conditions.[22,23] A good and significant correlation was found between a relative change in NB_{av} and in the ONH tissue blood flow rate determined with the hydrogen gas clearance method.[22,23] These results suggest that NB_{av} obtained from ONH also correlates with the ONH tissue blood flow rate, at least under certain conditions. NB_{av} obtained from the choroid showed a significant correlation with the choroidal blood flow rate determined using the microsphere technique in the same rabbit eyes[16,24] and NB_{av} obtained from a retinal area free of surface vessels in Dutch rabbits using the laser speckle method with the argon blue laser showed a significant correlation with the retinal blood flow rate determined using the microsphere technique in the same rabbit eyes.[20] These results also suggest that NB_{av} obtained from choroid or retina also correlates with the choroidal or retinal tissue blood flow rate, at least under certain conditions. In human eyes, the coefficients of reproducibility of 1 min and 24 h interval measurements of NB_{ONH} were 11.7% and 13.0%, and those of $NB_{ch\text{-}ret}$ were 8.7% and 9.7%, respectively.[19] Thus, because of good reproducibility of measurements from the same site and good correlation with the blood flow measurements with other methods, this method is very useful in following the time course of change in an individual tissue.

Unlike in rabbit eyes, in which the retinal vasculature exists only in the medullary wing area, the NB obtained from the posterior fundus in human eyes using the diode laser is affected by both the retinal and choroidal circulation.[25] NB_{av} obtained from the posterior fundus of monkeys using the diode laser before and after obstruction of a retinal artery (experimental branch retinal artery occlusion) suggested that the contribution of retinal microcirculation to NB_{av} was about 20% or less.[26] Results in monkey eyes suggest that the contribution of the choroidal circulation to the NB_{av} presently obtained in human eyes from a field located halfway between the macula and the ONH, with no discrete retinal vessels visible, is probably much greater than the contribution of the retinal microcirculation.

Effect of smoking on ONH and posterior fundus circulation

The results obtained in our testing of light smokers demonstrated that NB_{ONH} showed little difference and $NB_{ch\text{-}ret}$ a small but significant decrease after cigarette smoking, and that BP_m and PR significantly increased after smoking.

In the present testing of light smokers, the ocular perfusion pressure (OPP) calculated by the difference between $2/3BP_m$ and IOP should increase by about 20%, since BP_m significantly increased by about 10 mmHg between 1 and 20 min after smoking and the IOP was considered to have remained unchanged during the experiments. In spite of the increase in OPP, NB_{ONH} showed little change and $NB_{ch\text{-}ret}$ showed a small decrease after cigarette smoking. It is known that Poiseuille's law can be applied not only in the large vessels but also in arterioles.[27,28] According to the Poiseuille formula,[29] blood flow in the arteriole is given by

$$F = \pi/128 \times \Delta P \times 1/\eta \times D^4 \qquad (2)$$

where ΔP is the pressure difference along the vessel, η is the blood viscosity, and D is the vessel (blood column) diameter. Since the mean blood velocity, V_{mean}, is given by $F/(\pi D^2/4)$, V_{mean} can be calculated as follows:

$$V_{mean} = 1/32 \times \Delta P \times 1/\eta \times D^2 \qquad (3)$$

The term ΔP is thought to be increased by about 20%, since it roughly parallels the OPP, and V_{mean} (NB_{ONH} or $NB_{ch\text{-}ret}$) remained little changed or slightly decreased. Thus, D should have decreased by about 10%. The increase in BP_m after smoking was considered to be due to stimulation of the sympathetic nervous system and adrenal medulla by nicotine.[3,30,31] Therefore, the decrease in D in the ONH and choroid and/or retina may be at least partly attributable to the above mechanisms. Further, cigarette smoking or nicotine infusion increased plasma concentrations of vasopressin or angiotensin II[32–34] in addition to noradrenaline or adrenaline,[30,31] which might also lead to a larger decrease in D and decrease in blood flow. Previous reports have also demonstrated that smoking results in decreased blood flow in skin[4,33] and placenta,[5] which are compatible with the present result. With the present apparatus, the retinal and choroidal circulation cannot be measured separately and the contribution of the latter to $NB_{ch\text{-}ret}$ is thought to be much larger.[12] If we assume that vessel diameter in the choroidal vessels did not change after smoking in the present study, choroidal blood flow should have increased as the OPP increased because of its lack of autoregulation against OPP change,[16,35] which should result in a substantial increase in $NB_{ch\text{-}ret}$. Therefore, the small decrease in $NB_{ch\text{-}ret}$ obtained here strongly suggests that D in choroidal vessels should have decreased after smoking, which is possibly related to vasoconstriction due to catecholamines,[30] vasopressin or angiotensin II.[32–34]

Using the same laser speckle method, we have recently found that smoking increased tissue blood velocity in the ONH and possibly that in the choroid in habitual smokers whose smoking history was longer than 10 years and smoking index was 18 times that of the present subjects.[7] Taken together with our previous result, the present result is thought to indicate that the effect of smoking on the ONH or posterior fundus circulation is different between habitual smokers with a long smoking history and those with a short smoking history. As stated in the introduction, the effect of cigarette smoking on the tissue blood flow may be different between non-smokers and chronic smokers,[13–15] and an animal experiment suggested that this may be also the case for the choroidal blood flow.[11,12]

Habitual smoking is reported to reduce baseline cerebral blood flow by chronically raising resistance in cerebral blood vessels.[36] The absorption of nicotine should produce constriction of the peripheral vasculature thereby increasing the peripheral resistance to flow, while smoking leads to an increase in carbon monoxide which causes a reduction in oxygen transport into the tissues, and thereby downstream vasodilation and an increase in blood flow velocity.[37] The difference in the effect of cigarette smoking on the circulation between light and heavy smokers may be at least partly attributable to differences in the vascular bed reaction to the contrasting effects of the absorption of nicotine and carbon monoxide.

In summary, the present results indicated that cigarette smoking caused little or a small decrease in tissue blood velocity, but decreased peripheral vessel diameters in the ONH and probably in the choroid in young healthy light smokers. Further, the effect of cigarette smoking on these tissues in light smokers was suggested to differ from that in habitual heavy smokers.

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