PAPER

Bedside assessment of cerebral perfusion reductions in patients with acute ischaemic stroke by near-infrared spectroscopy and indocyanine green

C Terborg, S Bramer, S Harscher, M Simon, O W Witte

J Neurol Neurosurg Psychiatry 2004;**75**:38–42

See end of article for authors' affiliations

Correspondence to: Dr Christoph Terborg, Department of Neurology, Friedrich-Schiller University of Jena, Philosophenweg 3, 07740 Jena, Germany; christoph.terborg@med.uni-jena.de

Received 19 February 2003
In revised form 24 May 2003
Accepted 26 May 2003

Objective: To detect perfusion reductions in patients with acute cerebral infarcts using near-infrared spectroscopy (NIRS) with indocyanine green (ICG) as tracer.
Methods: Kinetics of an intravenous bolus of ICG were monitored by NIRS in 13 patients with acute infarction in the territory of the middle cerebral artery (mean (SD) age, 62.2 (13.0) years) and 12 controls (64.2 (9.1) years) at 2.8 (2.8) days after onset. NIRS optodes were placed bitemporally, with an interoptode distance of 4–5 cm. Absolute concentration changes in ICG were calculated. The following were assessed: time to peak, maximum ICG concentration, time interval between 0% and 100% maximum ICG concentration (interval), rise time (time between 10% and 90% ICG maximum), slope (maximum Δ ICG/interval), and blood flow index (BFI = maximum Δ ICG/rise time) of each hemisphere. Intraindividual differences were calculated between the two hemispheres.
Results: Patients with ischaemic stroke had increased time to peak ($p<0.01$), interval ($p<0.01$), and rise time ($p<0.01$), while maximum ICG concentration ($p<0.03$), slope ($p<0.01$), and BFI ($p<0.01$) were diminished at the site of infarction compared with the unaffected hemisphere. In stroke patients, intraindividual differences in time to peak ($p<0.001$), interval ($p<0.001$), rise time ($p=0.001$), maximum ICG concentration ($p<0.02$), slope ($p<0.001$), and BFI ($p<0.001$) were greater than in the controls, with excellent sensitivity and specificity for Δ time to peak (100% and 100%, respectively) and Δ time interval (100% and 91.7%).
Conclusions: Measurement of interhemispheric differences in ICG kinetics by NIRS detects perfusion reductions in patients with acute middle cerebral artery infarction. This non-invasive bedside test is rapid, repeatable, without major side effects, and avoids transportation of critically ill patients.

Assessment of regional cerebral blood flow (rCBF) might be useful in patients with acute ischaemic stroke, especially in relation to thrombolytic treatment. There are several neuroradiological techniques providing rCBF measurement: single photon emission tomography, xenon computed tomography (CT), positron emission tomography (PET), perfusion weighted magnetic resonance imaging (MRI), and recently dynamic single slice computed tomographic imaging. All these allow measurement of cerebral perfusion deficits. However, repeated measurements of cerebral blood flow require appropriate technical equipment, and transportation may be hazardous in the critically ill patient.

rCBF has recently been measured in pigs by monitoring the kinetics of the dye indocyanine green (ICG) by near infrared spectroscopy (NIRS), and the data correlated well with values assessed by radioactive microspheres.[1] The aim of our study was to detect cerebral perfusion reductions in patients with acute ischaemic stroke in the territory of the middle cerebral artery by a rapid bedside test using indocyanine green as a tracer and NIRS. We focused our interest on calculating time to peak, peak intensity, and slope, which were previously shown to correlate with cerebral perfusion deficits in acute stroke in CT and MRI studies.[2 3]

METHODS

We studied 13 patients (five female, eight male; mean (SD) age, 62.2 (13.0) years) with symptoms of acute middle cerebral artery ischaemia. The symptoms were confirmed by CT or MR tomography, which showed a stroke volume of between 50% and 100% in the territory of the middle cerebral artery, with or without infarction of the anterior cerebral artery (one case). The kinetics of an intravenous bolus of indocyanine green (ICG-Pulsion, Pulsion Medical Systems, Munich, Germany) were monitored by NIRS (Niro 300, Hamamatsu Photonics, Herrsching, Germany) at 2.8 (2.8) days (range 0 to 8) after symptom onset. All patients were evaluated on the National Institutes of Health stroke scale (NIHSS) score[4] at the time of measurement, or, in ventilated patients, on the last day before intubation.

Dosage of indocyanine green was 0.22 (0.19) mg/kg: 0.5 mg/kg in patients with cardiac monitoring (n = 4) and otherwise 0.1 mg/kg (n = 9). The agent was given through a central venous line, or into the antecubital vein through an 18 gauge intravenous catheter.

The NIRS optodes were placed bitemporally on the scalp above the territory of the middle cerebral artery, with an interoptode distance of 4 or 5 cm laterally. The spectrometer employed (NIRO 300) provides light in the near-infrared region at four distinct wavelengths (775, 810, 853, and 913), and the receiving probe collected the scattered light with a sample rate of 6 Hz.

Absolute concentration changes of indocyanine green were calculated from light attenuation according to a modified Beer-Lambert law using specific software (Hamamatsu Photonics). For each measurement, we assessed the time to

Abbreviations: BFI, blood flow index; ICG, indocyanine green; NIHSS, National Institutes of Health stroke scale; NIRS, near-infrared spectroscopy; rCBF, regional cerebral blood flow

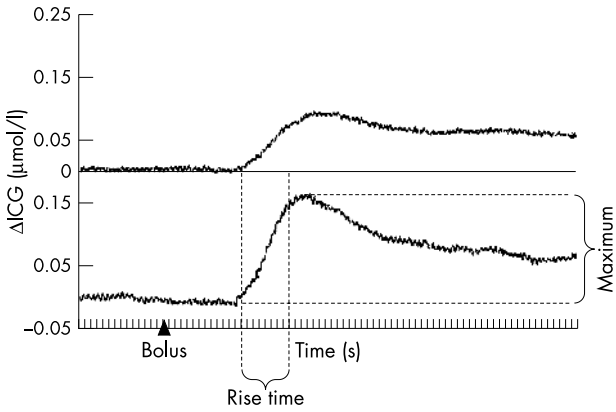

Figure 1 Typical indocyanine green (ICG) measurement in a patient with right middle cerebral artery infarction. Upper graph: cerebral ICG kinetics (right temporal); lower graph: ICG kinetics (left temporal) including calculation of blood flow index.

peak, the maximum indocyanine green concentration (μmol/l), the time interval between 0% and 100% of the maximum signal (interval), the rise time (defined as time between 10% and 90% of the ICG maximum), the slope (maximum Δ ICG/interval), and the blood flow index (maximum Δ ICG/rise time) of both hemispheres, as described before.[1] We also calculated the intraindividual difference in indocyanine green kinetics between the two hemispheres. A typical measurement is shown in fig 1.

Controls were 12 patients (aged 64.2 (9.1) years) under the care of the department of anaesthesiology and intensive care medicine in our hospital who did not have central nervous system disease. In these patients, indocyanine green was given in a dose of 0.5 mg/kg for cardiac monitoring or liver function tests. Significant carotid artery disease was excluded by Doppler sonography.

The protocol was approved by the local ethics committee. Informed consent was obtained from patients or their relatives before the procedure.

Statistics

As several values did not have a normal distribution, comparisons between patients with ischaemic infarction in the middle cerebral artery territory and the controls were done using a non-parametric Mann-Whitney U test. The intraindividual differences between the two hemispheres were calculated using a matched-pairs Wilcoxon test. The validity of ICG kinetic indices was assessed by sensitivity, specificity, and receiver operating characteristic (ROC) analysis. Statistical significance was assumed at the 5% level.

RESULTS

The clinical characteristics of the patients with ischaemic infarction, including their NIHSS scores and extracranial Doppler or Duplex sonography results, are shown in table 1.

Intraindividual comparisons

In patients with ischaemic stroke, time to peak, interval, and rise time were significantly increased at the site of infarction compared with the unaffected hemisphere, and the slope and blood flow index were reduced (Wilcoxon; $p<0.01$). The maximum indocyanine green concentration was slightly reduced in the ischaemic hemisphere ($p<0.03$). The results are summarised in table 2. We did not find any difference in indocyanine green kinetics between the two hemispheres in the control group (table 3).

Comparison between patients with middle cerebral artery infarction and controls

Patients with infarction in the territory of the middle cerebral artery had an increased time to peak, interval, and rise time

Table 2 Comparison of indocyanine green kinetics between the affected and the unaffected hemisphere in patients with ischaemic stroke (matched pairs Wilcoxon test)

Variable	MCA infarction	Unaffected hemisphere	Wilcoxon p Value
Time to peak (s)	25.6 (9.2)	20.5 (6.5)	<0.01
ICG peak (Δμmol/l)	0.087 (0.062)	0.102 (0.063)	<0.03
Time interval (0–100%) (s)	16.5 (9.2)	10.8 (4.1)	<0.01
Rise time (10–90%) (s)	10.6 (5.7)	7.0 (2.7)	<0.01
Slope (μmol/l/s)	0.0073 (0.0071)	0.0111 (0.0088)	<0.01
BFI (μmol/l/s)	0.0118 (0.0118)	0.0185 (0.0163)	<0.01

Values are mean (SD).
BFI, blood flow index; ICG, indocyanine green; MCA, middle cerebral artery.

Table 1 Clinical characteristics of patients with ischaemic infarction, with National Institutes of Health stroke scale scores and extracranial Doppler or duplex sonography results

Patient	Age (years), sex	NIHSS	Site of infarction	Ipsilateral internal carotid artery	Contralateral internal carotid artery
1	71, M	19*	MCA left	Occlusion	–
2	74, F	17*	MCA right	Occlusion (distal)	–
3	57, M	9*	MCA left	–	60% stenosis
4	57, M	17*	MCA left	Occlusion	Occlusion
5	79, F	23	MCA left	Occlusion (distal)	–
6	76, F	17	MCA left	–	–
7	73, F	17	MCA left	–	–
8	44, F	18	MCA left	–	–
9	61, M	14*	MCA right	Occlusion	–
10	73, M	12*	MCA right	Occlusion	–
11	41, M	17	MCA left	Dissection	–
12	47, M	19*	MCA left	–	–
13	55, M	19*	ACA+MCA left	Occlusion	–

*Assessed before ventilation.
ACA, anterior cerebral artery; F, female; M, male; MCA, middle cerebral artery; NIHSS, National Institutes of Health stroke scale.

Table 3 Comparison of indocyanine green kinetics between the two hemispheres in controls (matched pairs Wilcoxon test)

Variable	Left hemisphere	Right hemisphere	Wilcoxon p Value
Time to peak (s)	18.2 (4.9)	18.6 (4.9)	>0.05
ICG peak (Δμmol/l)	0.194 (0.026)	0.188 (0.030)	>0.05
Time interval (0–100%) (s)	10.0 (2.7)	10.0 (2.5)	>0.05
Rise time (10–90%) (s)	6.6 (2.0)	6.7 (2.0)	>0.05
Slope (μmol/l/s)	0.0210 (0.0079)	0.0199 (0.0079)	>0.05
BFI (μmol/l/s)	0.0327 (0.0134)	0.0314 (0.0124)	>0.05

Values are mean (SD).
BFI, blood flow index; ICG, indocyanine green.

Table 4 Intraindividual differences of cerebral indocyanine green kinetics between the two hemispheres in patients with ischaemic stroke and controls (Mann-Whitney U test)

Variable	Patients with ischaemic stroke	Controls	p Value
Δ Time to peak (s)	5.2 (5.2)	−0.3 (0.6)	<0.001
Δ ICG peak (μmol/l)	−0.015 (0.019)	−0.006 (0.0168)	<0.02
Δ Time interval (s)	5.69 (6.82)	0.05 (0.50)	<0.001
Δ Rise time (s)	3.68 (4.27)	−0.10 (0.73)	0.001
Δ Slope (μmol/l/s)	−0.0039 (0.0034)	−0.0011 (0.0019)	<0.001
Δ BFI (μmol/l/s)	−0.0067 (0.0069)	−0.0011 (0.0019)	<0.001

Values are mean (SD).
BFI, blood flow index; ICG, indocyanine green.

in the affected hemisphere compared with the controls, but the difference was not significant. A comparison of maximum indocyanine green concentrations and maximum dependent indices (slope and blood flow index) could not be made between the two groups owing to the use of different ICG dosages (controls, 0.5 mg/kg; stroke patients, 0.22 (0.19) mg/kg; p<0.01).

The intraindividual differences of all parameters of cerebral indocyanine green kinetics between the two hemispheres in patients with ischaemic stroke differed significantly from those in the controls (table 4). The interhemispheric differences in time to peak, interval, and blood flow index in controls and index cases are shown in figs 2–4.

Sensitivity and specificity of the intraindividual differences of cerebral ICG kinetics were: Δ time to peak, 100% and 100% (p = 0.000); Δ maximum ICG concentration, 69.2% and 66.7% (p = 0.012); Δ time interval, 100% and 91.7% (p = 0.000); Δ rise time, 84.6% and 83.3% (p = 0.001); Δ slope, 84.6% and 91.7% (p = 0.000); and Δ BFI, 76.9% and 91.7% (p = 0.000; table 5).

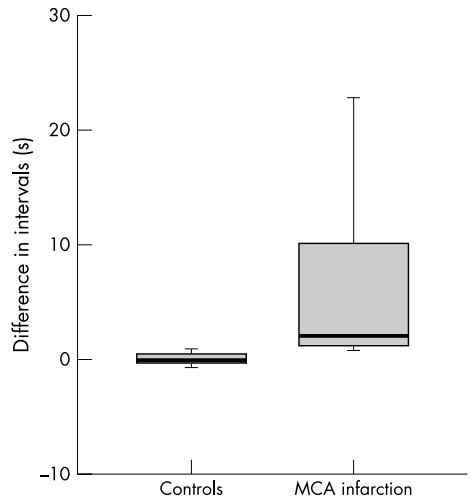

Figure 3 Interhemispheric difference in interval (between 0% and 100% of the maximum signal) in controls and patients with middle cerebral artery infarction (Mann-Whitney U test, p<0.001). MCA, middle cerebral artery.

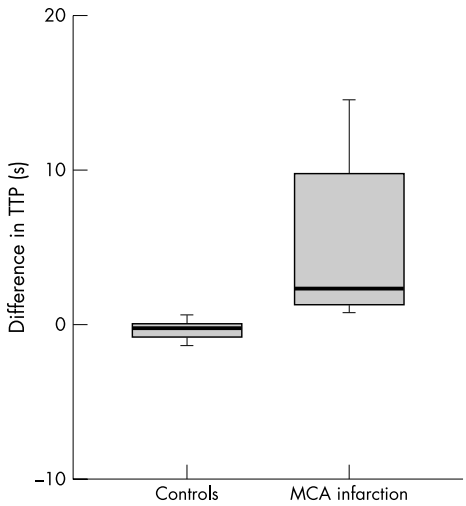

Figure 2 Interhemispheric difference in time to peak (TTP) in controls and patients with middle cerebral artery infarction (Mann-Whitney U test, p<0.001). MCA, middle cerebral artery.

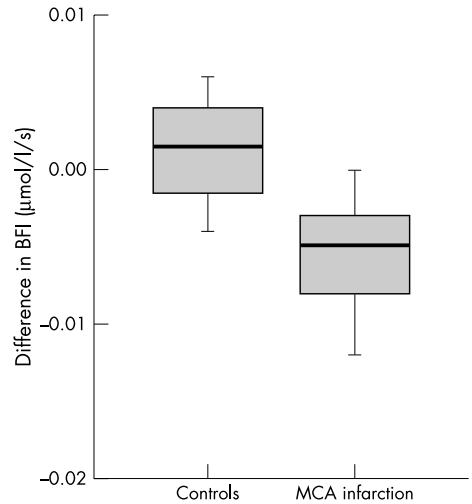

Figure 4 Interhemispheric difference in blood flow index (BFI, = maximum ΔICG/rise time) in controls and patients with middle cerebral artery infarction (Mann-Whitney U test: p<0.001). ICG, indocyanine green; MCA, middle cerebral artery.

Table 5 Sensitivity, specificity, and receiver operating characteristic analysis of interhemispheric differences in indocyanine green kinetics

Variable	Sensitivity (%)	Specificity (%)	Threshold value	Area under the curve	p Value
Δ Time to peak	100	100	0.74 s	1.0	0.000
Δ ICG peak	69.2	66.7	−0.005 μmol/l	0.795	0.012
Δ Time interval	100	91.7	0.74 s	0.994	0.000
Δ Rise time	84.6	83.3	0.58 s	0.881	0.001
Δ Slope	84.6	91.7	−0.0015 μmol/l/s	0.955	0.000
Δ BFI	76.9	91.7	−0.0025 μmol/l/s	0.913	0.000

BFI, blood flow index; ICG, indocyanine green.

DISCUSSION

Perfusion deficits in acute stroke can be detected by CT or MRI with high spatial resolution,[2 5–7] but these measurements require elaborate technical equipment and transportation. This might not be feasible in critically ill patients, especially if follow up measurements are needed. Thus innovative techniques for bedside measurement of cerebral perfusion are required.

We assessed reduction in cerebral perfusion in patients with clinically and radiologically confirmed acute middle cerebral artery infarction non-invasively using NIRS with indocyanine green as a tracer. While an interindividual comparison of indocyanine green kinetics between patients and controls was complicated by different dosages, administration routes, and possible variabilities in cardiac output, calculation of intraindividual differences eliminates these shortcomings. The intraindividual differences in all variables were significantly greater in patients with middle cerebral artery infarction than in the controls (table 4), and ROC analysis showed excellent sensitivity and specificity for the parameters Δ time to peak and Δ time interval (table 5). This method might therefore enhance signal to noise ratio in patients with hemispheric stroke.

Several studies have involved the non-invasive assessment of rCBF by NIRS with oxyhaemoglobin as the intravascular tracer after rapid changes in oxygen saturation.[8–10] In neonates, measurements in the transmission mode correlated well with the ^{133}xenon clearance.[11] However, this method was shown to be inaccurate in the reflectance mode and therefore cannot be applied in adults.[12]

Assessment of cerebral haemodynamics by NIRS with an intravenous indocyanine green bolus yields time–intensity curves of cerebral perfusion independent of oxyhaemoglobin.[10 13] In newborn infants, rCBF values using indocyanine green correlated well with those using oxyhaemoglobin as a tracer.[14] However, up to now the absolute quantification of rCBF by NIRS with indocyanine green has proved difficult owing to the unknown optical path length of photons through biological tissue, scattering losses, the influence of extracerebral tissue, and the need for an arterial input function to apply Fick's principle.[1] Cerebral blood volume has been measured in healthy subjects by NIRS and indocyanine green using an integration method, but values were substantially lower than those obtained using single photon emission computed tomography (SPECT) or PET, probably because of the above mentioned methodological limitations.[15] Recently, precise measurements of cerebral blood flow have been obtained from a bolus passage of indocyanine green using Fick's principle in the differential form in newborn piglets,[16] but until now commercial devices do not provide for the determination of differential path lengths. However, assessment of rCBF in animals by indocyanine green kinetics showed a strong correlation with rCBF measured by radioactive microspheres.[1 10]

Skin blood flow has been found to contaminate cerebral NIRS measurements,[17] but its influence on indocyanine green kinetics seems to be limited. In a porcine model, Kübler *et al* did not find a correlation between blood flow index and skin blood flow, but they did with rCBF.[1] Interestingly, measurement of rCBF derives from the first part of the time–intensity curve up to the peak. This corresponds to recent time of flight and frequency domain studies,[18–20] proving that an intravenous indocyanine green bolus arrives early in cerebral tissue and is delayed in the upper layers. This means that the first part of the curve used for determining indocyanine green kinetics mainly represents rCBF and is less contaminated by extracerebral tissue, especially skin blood flow. Accordingly, our results of perfusion reductions restricted to the site of infarction cannot be explained by extracerebral contributions, particularly as significant stenoses of the external carotid arteries were excluded by Doppler and duplex sonography.

Recently, global cerebral blood flow was measured using indocyanine green by a double indicator dilution technique (thermodilution and dye dilution curve), with good agreement with the Kety-Schmidt method, and its application in patients with space occupying middle cerebral artery infarction correlated well with outcome.[21 22] However, this method requires catheterisation of the jugular bulb and the femoral artery and does not provide regional resolution. It may thus not detect rCBF changes.

Conclusions

Our study shows that measurement of indocyanine green kinetics by means of NIRS (in particular Δ time to peak) provides a useful tool for the detection of reduced perfusion in patients with middle cerebral artery infarction. Assessment of asymmetries in indocyanine green kinetics in acute stroke does not substitute for cerebral imaging by CT or MRI before thrombolysis, but may be important in monitoring the effect of thrombolysis, as well as during carotid endarterectomy to detect embolic complications, and even in situations when sequential neurological examinations cannot be done in sedated patients. The method is non-invasive, repeatable, without major side effects, and can easily be done at the bedside. Although absolute quantification of rCBF at the bedside has not proved possible up to now, the value of repeated measurements of cerebral perfusion in intensive care units and during cardiac surgery should be investigated, in order to optimise blood pressure and cardiac output in order to avoid global cerebral hypoperfusion. Furthermore, developments in multichannel NIRS are yielding topographical information on cerebral perfusion with the use of indocyanine green kinetics, and this may have important implications for improved spatial resolution and for assessing different cerebrovascular territories.[13]

Authors' affiliations
C Terborg, S Bramer, S Harscher, O W Witte, Department of Neurology, Friedrich-Schiller University of Jena, Jena, Germany
M Simon, Department of Anaesthesiology and Intensive Care, Friedrich-Schiller University of Jena

Competing interests: none declared

REFERENCES

1 **Kübler WM**, Sckell A, Habler O, *et al*. Noninvasive measurement of regional cerebral blood flow by near-infrared spectroscopy and indocyanine green. *J Cereb Blood Flow Metab* 1998;**18**:445–56.
2 **Röther J**, Jonetz Mentzel L, Fiala A, *et al*. Hemodynamic assessment of acute stroke using dynamic single-slice computed tomographic perfusion imaging. *Arch Neurol* 2000;**57**:1161–6.
3 **Grandin CB**, Duprez TP, Smith AM, *et al*. Which MR-derived perfusion parameters are the best predictors of infarct growth in hyperacute stroke? Comparative study between relative and quantitative measurements. *Radiology* 2002;**223**:361–70.
4 **Lyden P**, Brott T, Tilley B, *et al*. Improved reliability of the NIH Stroke Scale using video training. NINDS TPA Stroke Study Group. *Stroke* 1994;**25**:2220–6.
5 **Reichenbach JR**, Röther J, Jonetz Mentzel L, *et al*. Acute stroke evaluated by time-to-peak mapping during initial and early follow-up perfusion CT studies. *Am J Neuroradiol* 1999;**20**:1842–50.
6 **Tong DC**, Yenari MA, Albers GW, *et al*. Correlation of perfusion- and diffusion-weighted MRI with NIHSS score in acute (<6.5 hour) ischemic stroke. *Neurology* 1998;**50**:864–70.
7 **Sorensen AG**, Buonanno FS, Gonzalez RG, *et al*. Hyperacute stroke: evaluation with combined multisection diffusion-weighted and hemodynamically weighted echo-planar MR imaging. *Radiology* 1996;**199**:391–401.
8 **Edwards D**, Wyatt JS, Richardson C, *et al*. Cotside measurements of cerebral blood flow in ill newborn infants by near infrared spectroscopy. *Lancet* 1988;ii:770–1.
9 **Owen Reece H**, Elwell CE, Harkness W, *et al*. Use of near infrared spectroscopy to estimate cerebral blood flow in conscious and anaesthetized adult subjects. *Br J Anaesth* 1996;**76**:43–8.
10 **Roberts IG**, Fallon P, Kirkham FJ, *et al*. Measurement of cerebral blood flow during cardiopulmonary bypass with near-infrared spectroscopy. *J Thorac Cardiovasc Surg* 1998;**115**:94–102.
11 **Bucher HU**, Edwards AD, Lipp AE, *et al*. Comparison between near infrared spectroscopy and ^{133}xenon clearance for estimation of cerebral blood flow in critically ill preterm infants. *Pediatr Res* 1992;**33**:56–60.
12 **Newton CR**, Wilson DA, Gunnoe E, *et al*. Measurement of cerebral blood flow in dogs with near infrared spectroscopy in the reflectance mode is invalid. *J Cereb Blood Flow Metab* 1997;**17**:695–703.
13 **Kusaka T**, Isobe K, Nagano K, *et al*. Estimation of regional cerebral blood flow distribution in infants by near-infrared topography using indocyanine green. *Neuroimage* 2001;**13**:944–52.
14 **Patel J**, Marks K, Roberts I, *et al*. Measurement of cerebral blood flow in newborn infants using near infrared spectroscopy with indocyanine green. *Pediatr Res* 1998;**43**:34–9.
15 **Hopton P**, Walsh TS, Lee A. Measurement of cerebral blood volume using near-infrared spectroscopy and indocyanine green elimination. *J Appl Physiol* 1999;**87**:1981–7.
16 **Springett R**, Sakata Y, Delpy DT. Precise measurement of cerebral blood flow in newborn piglets from the bolus passage of indocyanine green. *Phys Med Biol* 2001;**46**:2209–25.
17 **Smielewski P**, Czosnyka M, Pickard JD, *et al*. Clinical evaluation of near-infrared spectroscopy for testing cerebrovascular reactivity in patients with carotid artery disease. *Stroke* 1997;**28**:331–8.
18 **Steinbrink J**, Wabnitz H, Rinneberg H, *et al*. Determining changes in NIR absorption using a layered model of the human head. *OSA Technical Digest* 2000:278–80.
19 **Steinbrink J**, Wabnitz H, Obrig H, *et al*. Determining changes in NIR absorption using a layered model of the human head. *Phys Med Biol* 2001;**46**:879–96.
20 **Kohl-Bareis M**, Obrig H, Steinbrink J, *et al*. Non-invasive monitoring of cerebral blood flow by a dye bolus method: separation of brain from skin and skull signals. *J Biomed Opt* 2002;**7**:464–70.
21 **Wietasch GJ**, Mielck F, Scholz M, *et al*. Bedside assessment of cerebral blood flow by double-indicator dilution technique. *Anesthesiology* 2000;**92**:367–75.
22 **Keller E**, Wietasch G, Ringleb P, *et al*. Bedside monitoring of cerebral blood flow in patients with acute hemispheric stroke. *Crit Care Med* 2000;**28**:511–16.

ECHO

Myotonic dystrophy like mitochondrial myopathy with tRNA^{Ala} mutation

Please visit the *Journal of Neurology, Neurosurgery, and Psychiatry* website [www.jnnp.com] for a link to the full text of this article.

In the past decade there have been reports of over 100 mitochondrial (mt) DNA mutations associated with neuromuscular disease. These have included large scale mtDNA rearrangements, common point mutations in mt tRNA genes, and mutations affecting structural genes of mtDNA encoded respiratory chain subunits. The tRNA mutation of MELAS (mitochondrial encephalomyopathy, lactic acidosis, and stroke-like episodes) is located in the $tRNA^{Leu\ (UUR)}$ gene and another 13 pathogenic mutations associated with neurodegenerative disorders have been described in this gene. Mutations in $tRNA^{Ile}$, $tRNA^{Lys}$, and $tRNA^{Ser(UCN)}$ have been associated with encephalomyopathies or non-syndromic deafness. A $tRNA^{Ala}$ mutation has been described in a patient with late onset progressive external ophthalmoplegia and dysphagia. A different heteroplasmic G>A mutation at position 5650 of mt $tRNA^{Ala}$ was reported in a 53 year old patient with symptoms of cerebral autosomal dominant arteriopathy with subcortical infarcts and leucoencephalopathy (CADASIL) together with a myopathy with numerous ragged red fibres. Now workers in Germany have described a patient with a mitochondrial myopathy and features of muscular dystrophy who has a heteroplasmic G5650 mutation in the mtDNA encoded $tRNA^{Ala}$ gene.

The patient's mother died of a stroke at the age of 36. He was normal up to the age of 14 when he developed weakness on exercise and difficulty climbing stairs. There was slow progression of proximal muscle weakness and atrophy in the legs and proximal arm weakness was first noticed at the age of 32. Muscle biopsy at age 36 showed appearances of chronic myopathy and 5–10% of fibres had ragged red and/or ragged blue appearance, indicating a mixed degenerative myopathy and mitochondrial disorder. EMG showed myotonic discharges. On electron microscopy there was marked mitochondrial proliferation with structural abnormalities of mitochondria. Screening of the entire mtDNA revealed a heteroplasmic mutation in $tRNA^{Ala}$ at np5650. The mutation was concentrated particularly in ragged red fibres. The rate of apoptosis in muscle cells was not increased.

▲ *Journal of Medical Genetics* 2003;**40**:752–757.