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Prevalence of HIV-1 drug resistance in treated patients with viral load >50 copies/mL: a 2014 French nationwide study

L. Assoumou[1], C. Charpentier[2], P. Recordon-Pinson[3], M. Grudé[1], C. Pallier[4], L. Morand-Joubert[5], S. Fafi-Kremer[6], A. Krivine[7], B. Montes[8], V. Ferré[9], M. Bouvier-Alias[10], J.-C. Plantier[11], J. Izopet[12], M.-A. Trabaud[13], S. Yerly[14], J. Dufayard[15], C. Alloui[16], L. Courdavault[17], H. Le Guillou-Guillemette[18], A. Maillard[19], C. Amiel[20], A. Vabret[21], C. Roussel[22], S. Vallet[23], J. Guinard[24], A. Mirand[25], A. Beby-Defaux[26], F. Barin[27], A. Allardet-Servent[28], R. Ait-Namane[1], M. Wirden[29], C. Delaugerre[30], V. Calvez[29], M.-L. Chaix[30], D. Descamps[2] and S. Reigadas[3,31]* on behalf of the ANRS AC-11 Resistance Study Group†

[1]Sorbonne Universités, UPMC Univ. Paris 06, INSERM, Institut Pierre Louis d'Épidémiologie et de Santé Publique (IPLESP UMRS 1136), F75013 Paris, France; [2]INSERM UMR1137, IAME Université Paris Diderot Sorbonne Paris Cité, AP-HP, Laboratoire de Virologie, Hôpital Bichat-Claude Bernard, Laboratoire Associé au Centre National de Référence du VIH-Résistance aux Antirétroviraux, Paris, France; [3]PTBM, Laboratoire de Virologie, Hôpital Pellegrin, CHU de Bordeaux; UMR 5234 MFP CNRS, Université de Bordeaux, 33076 Bordeaux cedex, France; [4]HU Paris sud, Hôpital Paul Brousse, Laboratoire de Virologie, Villejuif, France; [5]Sorbonne Universités, UPMC Univ. Paris 06, INSERM, Institut Pierre Louis d'Épidémiologie et de Santé Publique (IPLESP UMRS 1136), AP-HP, Laboratoire de Virologie, Hôpital Saint-Antoine, F75012 Paris, France; [6]Laboratoire de Virologie, Hôpitaux Universitaires de Strasbourg, Strasbourg, France; [7]AP-HP, Hôpital Cochin, Laboratoire de Virologie, Paris, France; [8]Laboratoire de Virologie, Hôpital Saint-Eloi, CHU Montpellier, Montpellier, France; [9]EA 4271, Nantes Université UFR Pharmacie, Laboratoire de Virologie, CHU Nantes, Nantes, France; [10]INSERM U955, National Reference Center for Viral Hepatitis B, C et Delta, Department of Virology, Henri Mondor Hospital, University of Paris-Est, Créteil, France; [11]Laboratoire de Virologie et COREVIH Haute-Normandie, CHU de Rouen, Rouen, France; [12]Laboratoire de Virologie, Hôpital Purpan de Toulouse, Toulouse, France; [13]Laboratoire de Virologie, Hôpital de la Croix-Rousse, Hospices Civils de Lyon, Lyon, France; [14]Laboratoire de Virologie, Hôpitaux Universitaires de Genève, Genève, Switzerland; [15]Laboratoire de Virologie, Hôpital l'Archet de Nice, Nice, France; [16]Laboratoire de Virologie, Hôpital Avicenne, APHP, HU Paris Seine Saint Denis, Bobigny, France; [17]Laboratoire de Virologie, Centre Hospitalier Victor Dupouy d'Argenteuil, Argenteuil, France; [18]Laboratoire de Virologie, CHU Angers et HIFIH Laboratory, UPRES 3859, SFR 4208, LUNAM University, Angers, France; [19]Laboratoire de Virologie, CHU de Rennes, Rennes, France; [20]AP-HP, Hôpital Tenon, Laboratoire de Virologie, Paris, France; [21]Laboratoire de Virologie, CHU Caen, Caen, France; [22]Laboratoire de Virologie, CHU Amiens, Amiens, France; [23]Laboratoire de Virologie, CHU Brest, Brest, France; [24]Laboratoire de Virologie, CHR Orléans, Orléans, France; [25]Laboratoire de Virologie, CHU Clermont-Ferrand, Clermont-Ferrand, France; [26]Laboratoire de Virologie, CHU Poitiers, Poitiers, France; [27]Laboratoire de Virologie, CHU Bretonneau, & INSERM U966, Tours, France; [28]Laboratoire de Virologie, CHU de Nimes, Nimes, France; [29]Sorbonne Universités, UPMC Univ. Paris 06, INSERM, Institut Pierre Louis d'épidémiologie et de Santé Publique (IPLESP UMRS 1136), AP-HP, Laboratoire de Virologie, Hôpital Pitié-Salpêtrière, F75013 Paris, France; [30]Laboratoire de Virologie, AP-HP, Hôpital Saint Louis, INSERM U941, Université Paris Diderot, Paris, France; [31]CRB plurithématique, Bordeaux Biothèques Santé, Groupe hospitalier Pellegrin-CHU de Bordeaux, Bordeaux, France

*Corresponding author. CRB Plurithématique, Bordeaux Biothèques Santé, Groupe Hospitalier Pellegrin-CHU de Bordeaux, Bordeaux, France.
Tel: +33-5-57-82-14-66; Fax: +33-5-57-82-15-12; E-mail: sandrine.reigadas@chu-bordeaux.fr
†Members are listed in the Acknowledgements section.

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Background: Surveillance of HIV-1 resistance in treated patients with a detectable viral load (VL) is important to monitor, in order to assess the risk of spread of resistant viruses and to determine the proportion of patients who need new antiretroviral drugs with minimal cross-resistance.

Methods: The HIV-1 protease and reverse transcriptase (RT) and integrase genes were sequenced in plasma samples from 782 consecutive patients on failing antiretroviral regimens, seen in 37 specialized centres in 2014. The genotyping results were interpreted using the ANRS v24 algorithm. Prevalence rates were compared with those obtained during a similar survey conducted in 2009.

Results: The protease and RT sequences were obtained in 566 patients, and the integrase sequence in 382 patients. Sequencing was successful in 60%, 78%, 78% and 87% of patients with VLs of 51–200, 201–500, 501–1000 and >1000 copies/mL, respectively. Resistance to at least one antiretroviral drug was detected in

56.3% of samples. Respectively, 3.9%, 8.7%, 1.5% and 3.4% of patients harboured viruses that were resistant to any NRTI, NNRTI, PI and integrase inhibitor (INI). Resistance rates were lower in 2014 than in 2009. Resistance was detected in 48.5% of samples from patients with a VL between 51 and 200 copies/mL.

Conclusion: In France in 2014, 90.0% of patients in AIDS care centres were receiving antiretroviral drugs and 12.0% of them had VLs >50 copies/mL. Therefore, this study suggests that 6.7% of treated patients in France might transmit resistant strains. Resistance testing may be warranted in all treated patients with VL > 50 copies/mL.

Introduction

Surveillance of HIV resistance ongoing in France for >10 years[1,2] in treated patients with detectable viral load (VL) is important to assess the risk of spread of resistant viruses and to determine the proportion of patients needing new drugs. Our aims were to describe mutations associated with virological failure (VF) in 2014 and to compare them with those found in a similar survey conducted in 2009. We also evaluated genotyping success rate according to plasma VL.

Patients and methods

Study population and data collection

This prospective, multicentre, cross-sectional study enrolled HIV-1-infected patients receiving combination ART (cART) for at least 6 months with two consecutive VL values of >50 copies/mL between September and December 2014. Sociodemographic and clinical data, treatment history and regimen at the time of VF, and HIV-1 resistance sequencing data were collected.

Ethics

The study was approved by an ethics committee (Comite Consultatif de Traitement de l'Information dans la Recherche Scientifique et Medicale) and by the national data confidentiality watchdog organization (Commission Nationale Informatique et Libertes), in keeping with French law. The patients received full information on their participation in the study and did not oppose the use of their data.

Genotypic resistance analysis

Protease, reverse transcriptase and integrase gene mutations were identified from the International AIDS Society resistance testing USA panel (version July 2014). Genotyping results were interpreted with the ANRS algorithm (2014, version 24). We considered that a virus was resistant to a drug when it was ranked 'resistant' or 'possibly resistant' to the drug.

Statistical analysis

Rates of successful sequencing and resistance were calculated according to plasma HIV RNA levels (51–200, 201–500, 501–1000 and >1000 copies/mL). Univariate and multivariable logistic regression models were used to identify factors predictive of the risk of amplification failure. Weighted analyses, based on number of patients from each centre, were used to derive representative estimates of percentages of patients harbouring viruses with resistance-associated mutations (RAMs). Virus was considered resistant to a class of drugs when ranked 'resistant' to all drugs in the class. We used the FHDH ANRS CO4 cohort dataset to estimate the percentage of treated patients with VL >50 copies/mL, who could potentially transmit resistant viruses.

Sequences from our previous 2009 survey[2] were re-analysed concomitantly to compare frequencies of RAMs between the two surveys. Multivariable logistic regression analysis was used to study the impact of period adjusted for factors that differed significantly between the two surveys.

Results

Characteristics of the study population

We included 782 patients in 37 centres. Protease and reverse transcriptase genes were successfully amplified in 566 (72.4%) patients. Amplification of integrase was successful in 382 of 480 cases.

The characteristics of the study population are summarized in Table 1. Patients were exposed to a median of 6 antiretroviral drugs (IQR 3–9), including 2 NRTIs (IQR 0–4), 1 NNRTI (IQR 0–1), 3 PIs (IQR 2–4) and 0 INIs (IQR 0–1). Ten percent were on first-line treatment.

Genotyping success rate

Success of resistance genotyping was 72.4% overall, and 87%, 78%, 78% and 60% for samples with VL >1000, 501–1000, 201–500 and 51–200 copies/mL, respectively. Multivariable analysis showed that risk of unsuccessful resistance genotyping was 4.7-fold higher (95% CI 3.0–7.2, $P < 0.001$) and 1.8-fold higher (95% CI 1.0–3.2, $P = 0.040$) when VL was 51–200 and 201–500 copies/mL, respectively, than when it was >1000 copies/mL. This risk was 2.5-fold higher (95% CI 1.7–3.5, $P < 0.001$) in cases of non-B subtype.

Genotypic resistance patterns

The most commonly observed NRTI RAMs were M184V/I (25.9%), and the most common thymidine analogue mutations were M41L, D67N, K70R, L210W, T215Y/F and K219Q/E (5.7%–13.5%). K65R and L74V were present in 2.1% and 3.6% of samples, respectively. The most frequent NNRTI RAMs were K103N (12.6%), Y181C (10.7%) and E138A/K (8.2%). Prevalence of major PI resistance mutations ranged from 0.2% (G48V) to 12.5% (V82A/F/M/S/T). The most common INI RAMs were N155H (5.2%), Q148H (2.9%), E157Q (3.4%) and G140S (2.4%). Q148H/R/K was preferentially associated with G140S (1.4%). R263K mutation was observed in 0.5% of samples.

Drug resistance interpretation

Resistance to at least one antiretroviral drug was observed in 56.3% of samples (at least one NRTI in 36.0%, NNRTI in 32.1%, PI in 20.2% and INI in 12.0%), and was less frequent in first-line treatment failure than in patients with multiple failures (42.1% versus 57.9%, $P = 0.025$). Percentages of viruses not susceptible to any antiretroviral drugs in the NRTI, NNRTI, PI and INI classes were 3.5%, 9.2%, 1.6% and 3.4%, respectively. Frequency of resistance

Table 1. Clinical and biological characteristics of HIV-1-infected patients on failing regimens included in the 2009 and 2014 surveys

	2009 ($n = 506$)	2014 ($n = 566$)	P
Age (years), median (IQR)	45 (39–51)	48 (39–54)	0.001
Gender, %			
Male	63	64	
Female	37	36	
Transmission group, %			
MSM	26	26	1.000
Other	74	74	
VL (log_{10} copies/mL), median (IQR)	3.0 (2.3–3.9)	2.8 (2.2–3.7)	<0.001
HIV RNA copies/mL, %			<0.001
51–200	19.4	36.4	
201–500	16.8	16.8	
501–1000	12.7	10.4	
>1000	51.2	36.4	
CD4 count (cells/mm^3), median (IQR)	335 (214–525)	375 (203–575)	0.429
HIV-1 subtype, %			0.001
B	61	51	
CRF02-AG	19	23	
HIV-1 CCR5 tropism ($n = 308$), %		66	
Time since HIV-1 diagnosis (years), median (IQR)	14 (7–19)	14 (721)	0.403
Time on ART (years), median (IQR)	10.7 (5–14)	11.1 (5–17)	0.002

to all members of at least one drug class was 13.4%. Resistance to all drugs in one, two, three and four antiretroviral drug classes was 11.1%, 1.2%, 0.8% and 0.3%, respectively.

In France in 2014, 90.0% of patients were receiving antiretroviral drugs and 12.0% of them had detectable VL. Therefore, although 56.3% of treated patients with detectable VL harboured viruses with resistance to at least one antiretroviral drug, extrapolation to the entire French database suggests that only 6.7% of treated patients would be at risk of transmitting resistant viruses.

Drug resistance according to VL

Resistance to at least one drug rose significantly with plasma VL ($P < 0.001$) (Figure 1). Resistance to at least one NRTI was more frequent at >200 copies/mL than at 51–200 copies/mL (38.9% versus 30.1%, $P = 0.036$), while this was not observed with other classes.

Comparison between 2009 and 2014 studies

VL at the time of VF was significantly lower in 2014 than in 2009 ($P < 0.001$) (Table 1).

Resistance was less prevalent in 2014 than in 2009. Multivariable analysis showed that the decrease in resistance rates was not influenced by factors that differed significantly between the two surveys (adjusted OR = 1.6, 95% CI 1.2–2.1, $P = 0.005$). This decline was not significant for nevirapine ($P = 0.051$) or efavirenz ($P = 0.080$). Prevalence of resistance to rilpivirine was higher in 2014 than in 2009 (20% versus 16%, $P = 0.020$) and stable for etravirine (12% versus 9%, $P = 0.166$).

Resistance to at least one drug was significantly less frequent in 2014 than in 2009 at the VL levels of 501–1000 copies/mL (61.0% versus 78.1%, $P = 0.039$) and >1000 copies/mL (57.8% versus 69.5%, $P = 0.009$). No difference was observed at VL levels of 51–200 and 201–500 copies/mL (Figure 1).

Discussion

We evaluated prevalence of RAMs among a large number of patients experiencing VF. Genotyping success was 72% overall, compared with 67% in the previous 2009 survey ($P = 0.023$).[2] This improvement was noteworthy among patients with low VL (<200 copies/mL: 34% versus 60%, $P < 0.001$) and might be due to more efficient amplification, as genotyping procedure changed between the two surveys. Our findings agree with those of another recent study.[3] Nevertheless, our multivariable analysis showed that low VL remains associated with amplification failure, together with non-B subtype viruses and confirmed limits of genotyping for non-B strains.[4]

Prevalence of RAMs fell significantly between the two surveys, whereas frequency of mutations E138A/K and H221Y conferring resistance to new-generation NNRTIs increased significantly, probably because of widespread use of these drugs. Mutation K103N was observed in 12.6% of patients, similar to the 2009 survey, despite less use of first-generation NNRTIs in 2014. High prevalence of K103N might be due to NNRTI past exposure but might also occur because the replicative capacity of viruses harbouring this mutation is almost preserved.[5,6] To date, there is no other nationwide survey of HIV-1 resistance in patients with VF in other countries since our 2009 survey. Resistance to at least one antiretroviral drug was detected in 56.3% of samples, which is in keeping with another study.[7]

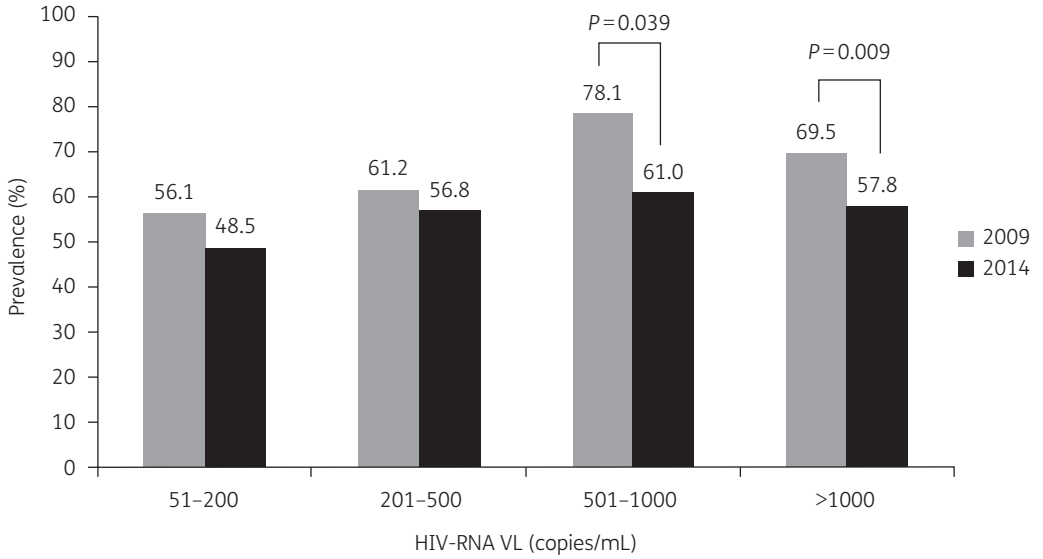

Figure 1. Resistance or possible resistance to at least one drug according to the HIV RNA level.

Prevalence of resistance to at least one antiretroviral drug was high (48.5%) among patients with low-level viraemia (LLV) of <200 copies/mL. Several studies have shown that LLV can promote the selection of resistance mutations.[8–10] Similar results to those obtained in our study showed that ∼50% of patients with LLV had viruses with RAMs.[3,11]

We found that prevalence of resistance to at least one PI, NNRTI or INI did not differ between patients with VL <200 copies/mL and those with VL >200 copies/mL. This supports the idea that genotyping must be attempted for all regimens, including those with low genetic barrier.

We found that resistance was less prevalent in 2014 than in 2009, except for next-generation NNRTIs. This decline may be explained by a reduction in use of older NNRTIs with a low genetic barrier. At the time of VF, 6.5% of patients were receiving etravirine in 2009, compared with 10.8% of patients in 2014 ($P = 0.012$); corresponding figures for rilpivirine were 0% and 7.4% ($P < 0.001$).

By contrast, among patients with LLV, we found no difference in resistance rates between 2009 and 2014 when stratifying on VL categories. This strongly supports guidelines recommending resistance monitoring for all treated patients with VL >50 copies/mL, even though low VL remains a risk factor for unsuccessful sequencing.

One limitation of this study is the lack of cumulative genotyping, which might underestimate the prevalence of resistance. Furthermore, the observed decline in prevalence of resistance between 2009 and 2014 is based on separate cross-sectional surveys of two different populations. Despite the lack of adherence data, ∼75% of patients had low VL (<1000 copies/mL), suggesting adherence to therapy.

In conclusion, in 2014, in France, 6.7% of treated patients might possibly transmit resistant strains, close to the percentage observed in naive patients.[12,13] Resistance was less prevalent than in 2009 except for new-generation NNRTIs. Our main findings are that resistance testing is feasible in most patients with VL <200 copies/mL and that resistance is evidenced in 50% of cases in patients with plasma VL <200 copies/mL. Therefore, resistance testing may be warranted, and largely feasible, in all treated patients with VL >50 copies/mL, challenging the definition of therapeutic failure in some current guidelines.

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Members of the ANRS AC-11 Resistance Study Group

Virology laboratories

Paris area. Avicenne: Chakib Alloui; Bichat: Diane Descamps, Charlotte Charpentier, Benoit Visseaux; Cochin: Anne Krivine; Henri Mondor: Magali Bouviers-Alias; Paul Brousse: Coralie Pallier; Pitié-Salpêtrière: Sidonie Lambert, Cathia Soulié, Marc Wirden, Anne-Geneviève Marcelin, Vincent Calvez; Saint-Antoine: Laurence Morand-Joubert, Djeneba Fofana; Saint-Louis: Nadia Mahjoub, Constance Delaugerre, Marie-Laure Chaix; Tenon: Corinne Amiel, Véronique Schneider.

Outside Paris area. Amiens: Catherine Roussel; Angers: Hélène Le Guillou-Guillemette; Argenteuil: Laurence Courdavault; Bordeaux: Sandrine Reigadas, Patricia Recordon-Pinson, Hervé Fleury; Brest: Sophie Vallet; Caen: Julia Dina, Astrid Vabret; Clermont-Ferrand: Audrey Mirand, Cécile Henquell; Dijon: Christelle Auvray, Alexis De Rougemont, Hélène Giraudon, Ali Si-Mohammed; Garches: Dominique Mathez; Grenoble: Anne Signori-Schmuck, Patrice Morand; Lille: Laurence Bocket; Lyon: Mary Anne Trabaud; Montpellier: Brigitte Montés; Nantes: Laura Le Guen, Audrey Rodallec, Virginie Ferre; Nancy: Hélène Jeulin, Evelyne Schvoerer; Nice: Jacqueline Dufayard; Nimes: Annick Allardet-Servent, Marie-Josée Carles; Orleans:

Jérôme Guinard, Aurélie Guigon; Poitiers: Geneviève Giraudeau, Agnès Beby-Defaux; Rennes: Anne Maillard; Rouen: Jean Christophe Plantier, Marie Leoz, Thomas Mourez; Saint-Etienne: Thomas Bourlet; Strasbourg: Samira Fafi-Kremer; Toulouse: Julie Chiabrando, Stéphanie Raymond, Jacques Izopet; Tours: Francis Barin; Versailles: Stéphanie Marque-Juillet.
Switzerland. Genève: Sabine Yerly.

Clinical centres

Paris area. Avicenne: Patricia Honoré; Bichat: Bao Phung, Sylvie Legac, Roland Landman, Yazdan Yazdanpanah; Cochin: Aline Maignan, Jean Paul Viard; Henri Mondor: Chrystel Chesnel, Jean-Daniel Lelievre, José-Luis Lopez-Zaragoza, Cécile Dumont; Jean Verdier: Sylvie Tassi; Paul Brousse: Claudine Bolliot, Martine Malet, Daniel Vittecoq; Pitié-Salpêtrière: Christine Blanc, Catherine Lupin, Anne Simon, Christine Katlama; Saint Antoine: Diane Bollens; Saint Louis: Diane Ponscarme, Jean-Michel Molina; Tenon: Valérie Berrebi, Gilles Pialloux.
Outside Paris area. Amiens: Nathalie Decaux, Jean-Luc Schmit; Angers: Eric Pichard, Sami Rehaiem, Pierre Abgueguen; Argenteuil: Juliette Gerbe, Philippe Genet; Bordeaux: Philippe Morlat, Marie-Anne Vandenhende, Fabrice Bonnet, Mojgan Hessamfar, Didier Neau, Michel Dupon, Charles Cazanave, Estibaliz Lazaro, Jean-Luc Pellegrin; Brest: Christopher Payan, Julien Boileau; Caen: Philippe Ferret; Clermont-Ferrand: Christine Jacomet; Dijon: Sandrine Gohier, Lionel Piroth; Garches: Huguette Berthe, P. De Truchis; Grenoble: Sylvie Dufresnes, Pascale Leclercq; Lille: Sylvie Vandamme; Lyon: Corinne Brochier, Djamila Makhloufi, Florence Garnier; Montpellier: Céline Fernandez, Jacques Reynes; Nantes: Hervé Hué, Laurence Larmet, Morane Cavellec, François Raffi; Nice: Aline Joulie, Pascal Pugliese; Nîmes: Regine Doncesco, Ganaelle Jacques, Albert Sotto; Orléans: Thierry Prazuck, Jennifer Buret, Laurent Hocqueloux, Catherine Mille, Mohamadou Niang; Poitiers: Gwenaël Le Moal, David Plainchamp; Rennes: Christian Michelet, Maja Ratajczak; Rouen: Francoise Borsa-Lebas, Francois Caron, Yasmine Debab; Saint-Etienne: Frédéric Lucht, Anne Frésard; Strasbourg: Patricia Fischer, Stéphanie Bertsch, David Rey; Toulouse: Florence Balsarin, Bruno Marchou; Tours: Frédéric Bastides, Louis Bernard, Guillaume Gras.
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Transparency declarations

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