Quist and Chapela reply — Our original publication[1] contained two separate conclusions derived from two methodological approaches. First, using PCR, we detected the presence of three distinct transgenic DNA sequences in maize landraces in Oaxaca, Mexico[1]. Second, we attempted to establish the genomic context of transgene insertion using i-PCR. The criticisms raised by Metz and Fütterer and by Kaplinsky *et al.* relate principally to our second statement.

In contrast with the well-established PCR method, i-PCR is an exploratory method that depends on interpretation and the availability of known sequences in databases such as GenBank. We acknowledge that our critics' assertion of the misidentification of sequences labelled with *adh1* intron 1 and with *bronze1* is valid.

The suggestion of mispriming in our i-PCR reaction is also warranted for sequences AF434756 and AF434759 (ref. 1). Significant homology with putative misamplifications is maintained across the length of these fragments, and the CaMV sequence was not recovered. However, this pattern is not found in our other i-PCR sequences. A revealing pattern of discontinuity is found at at least one end of five other sequences, indicating the integration junction between the transgenic DNA and the native host genome. Our critics choose not to recognize this feature in the majority of our i-PCR data. Partial homology with retrotransposon elements in maize is common in primers designed to amplify transposon-like sequences, and is not unique to our primers. Questions concerning the distortion of expected footprints at the DNA-integration junction certainly warrant future work.

The movement of transgenes into new populations and across generations is expected to result in diverse integration patterns[2–7]. Our findings are compatible with recent studies[2–6] that characterize transgene/host DNA junctions where rearrangements include interspersion with host or unidentifiable DNA. As altered DNA species should also be an important focus of ecological research, we disagree with our critics who assume that only intact transgenes are worthy of attention in our study.

We agree that PCR-based methods are sensitive and therefore open to artefacts, but strongly disagree that the presence of these artefacts is unavoidable or uncontrollable. The consistent performance of our controls, as reported[1], discounts beyond reasonable doubt the possibility of false positives in our results. Nevertheless, the high sensitivity of the PCR reaction has incited some critics to request a non-PCR-based method to confirm our main statement. To address these challenges, we evaluated the same samples from our original publication[1] using DNA–DNA hybridization. The results of these experiments continue to support our primary statement.

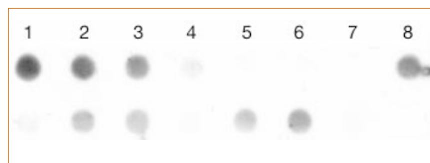

Figure 1 DNA–DNA dot-blot hybridization between maize genomic DNA and a CaMV p-35S probe. Sample numbers coincide with those in ref. 1. Top row: 1, 100% transgenic; 2, 10% transgenic; 3, 5% transgenic; 4, 1% transgenic, 5, 0.5% transgenic; 6, historical maize negative control; 7, water negative control; 8, Diconsa sample K1. Bottom row: 1, criollo sample B1; 2, criollo sample B2; 3, criollo sample B3; 4, criollo sample A1; 5, criollo sample A2; 6, criollo sample A3; 7, Peru maize negative control P1; 8, water negative control.

Our analysis of Oaxacan maize is unique for several reasons. First, we wished to document changes that occur within diverse populations of landraces (rather than single varieties or lines), for which no markers, restriction-enzyme digestion maps or linkage analyses have been developed. Second, we could not have predicted which (or how many) specific transgenic constructs (or derivatives) were present in the samples that we analysed. Third, our samples of ground, pooled kernels from individual maize cobs do not represent individual genomes. All of these factors render the application of DNA-hybridization methods difficult. To minimize confusion in interpreting the multiplicity of bands that would have been created by Southern hybridization with our samples, we chose to use dot blotting for our experiments.

We extracted genomic DNA from dry maize kernels[1]. Standards containing varying amounts of transgenic material were prepared by mixing flour from our positive control (Bt1) and our historical negative control[1]. We blotted and immobilized 10–15 μg of DNA from each sample onto a nylon membrane using a Bio-Dot apparatus (Bio-Rad). We generated a horseradish peroxidase-labelled DNA probe from the same 220-base-pair fragment of the p-35S CaMV promoter that was amplified from our previously reported[1] positive control (Bt1). Hybridization conditions were as follows: 56 °C, 6 ng ml^{-1} DNA probe, 1 hour. Washes were as follows: 3×5 min with $0.1\times$SSC/0.1% SDS at 56 °C, followed by 3×5 min with $2\times$SSC at room temperature. Loading homogeneity was confirmed by stripping and rehybridization of the experimental membrane with the 329-base-pair fragment from the maize-specific *zein* gene[1]. Probe labelling, hybridizations and detection were carried out using a North2South kit (Pierce Endogen), according to the manufacturer's specifications.

DNA from four of our six criollo landrace samples, and from the Diconsa sample, hybridized with our CaMV probe (Fig. 1). By using standardized mixtures of transgenic and non-transgenic maize, dot-blot hybridization suggests a ratio of transgenic to non-transgenic kernels in criollo cobs of the order of 1:100, as we had previously suggested[1] and as was confirmed by Mexican government studies[1]. This DNA-hybridization study confirms our original detection of transgenic DNA integrated into the genomes of local landraces in Oaxaca.

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correction

Reward value of attractiveness and gaze
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Reward-related responses have been registered in animal brains mostly in the ventral half of the striatum, from the nucleus accumbens to the pallidum. Considering the location of the response to attractive faces we describe, the observed activation was large and its spatial extent was not clear from Fig. 2, although we inferred that the ventral stratum was involved. From the plane shown, this activation more accurately extended ventrally into the striatum, specifically into the palladium; the nucleus accumbens proper was not activated. Dorsally, the activation extended into the anterior thalamus (as shown in Fig. 2). Our conclusions that the attractiveness of faces is processed in brain regions involved in evaluating the reward value of stimuli, and that this processing depends on gaze direction, are unaltered.

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