Comparing the Toxicity of Particulate Matter (PM) Collected by Different Samplers

Prepared for the Department for Environment, Food and Rural Affairs (DEFRA), the Scottish Executive, the Welsh Assembly Government and the DoE in Northern Ireland

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1 Background

Whilst the health effects attributable to long term exposures to elevated ambient particulate concentrations are well established, the toxicological basis for these effects are less well understood. The current scientific consensus suggests that the capacity of inhaled PM to elicit inflammation in the lung due to it oxidative properties may partially underlie the observed health effects (1,2). The capacity to cause oxidation reactions at the air-lung interface, or the particulate oxidative potential, reflects a number of particle characteristics, including composition, size and surface area. Therefore a single measure of oxidative potential would effectively integrate a large number of PM characteristics into a measure of direct biological relevance. Such an approach has been undertaken by Professor Kelly's group at King's College London utilising the reaction between ambient PM samples and physiological antioxidants to assess particulate oxidative potential (2). In this approach environmental PM are incubated in a synthetic respiratory tract lining fluid, the first physical interface encountered by inhaled PM in the lung, containing the three major water soluble antioxidants: ascorbate, urate and glutathione. By measuring the extent to which individual PM samples can deplete these antioxidants with time it is possible to arrive at a measure of oxidative potential, expressed as the % depletion of the individual antioxidants. Losses of glutathione and ascorbate in this model indicate the presence of 'redox-active' metal (3), and organic (quinone/hydroquinone) species associated with the particle (*4*). These losses have been attributed both by the capacity of these antioxidants to reduce metals and quinones/hydroquinones, as well as by their capacity to scavenge superoxide formed as a consequence of the reaction of these reduced metals and semi-quinone radicals with molecular oxygen (*5,6*). The redox potential of urate (*7*) however precludes its direct reduction of PM-associated metals and quinones. In addition, it is not a particularly good scavenger of superoxide radicals (*8*). It is however a highly effective scavenger of hydroxyl radicals (*8*), that are proposed to be formed during PM-induced oxidative injury, *in vitro* (*9*) and *in vivo* (*10*).

These determinations of PM oxidative-activity are however critically dependent on the PM sample used being representative of those breathed in ambient air. To date much of the work that has been undertaken has been based on PM extracted from standard TEOM filters and concern has been expressed regarding the possible losses of secondary and volatile organic species that may occur as a consequence of the 50^{o}C collection. To address whether this is a major concern we made use of the current PM Equivalence tests being undertaken by DEFRA at Teddington and Birmingham. This ongoing study permitted us to examine whether parallel filters collected at a given site had differing levels of oxidative activity depending on the temperature of collection, i.e. to determine whether losses in secondary species and volatile organics dramatically affected the measure PM oxidative potential. Parallel standard TEOM (50^{o}C) and FDMS (Filter Dynamic Measurement System) filters (30 and 4^{o}C) were collected as indicated above, PM extracted from their various filter extracts and their oxidative activity assessed in the synthetic RTLF model. All PM samples were compared on an equal mass basis.

Table 1: PM collections made at the Birmingham and Teddington sites

Instrument	Collection Condition
TEOM PM_{10}	Heated to 50ºC
FDMS PM_{10} TEOM Filter	Heated to 30ºC
FDMS PM_{10} Purge Filter	Chilled to 4ºC
FDMS $PM_{2.5}$ TEOM Filter	Heated to 30ºC
FDMS $PM_{2.5}$ Purge Filter	Chilled to 4ºC

2 Study Aim

In this study we examined the endogenous oxidant activity of ambient particles (PM_{10} and $PM_{2.5}$) collected in parallel at Birmingham and Teddington as part of the current DEFRA equivalence testing study.

The following specific questions were addressed:

- Is the oxidative activity of samples collected at 4ºC greater than that of PM samples collected at 50^{o}C (standard TEOM) consistent with the view that volatile PM components contribute significantly to PM oxidative activity?

- Whether there is a measurable difference in the oxidative potential of FDMS TEOM filter PM_{10} and $PM_{2.5}$ collected at 4 and 30^{o}C?

- Whether there is a site-specific difference in the oxidative potential between the two sites?

- As there are two co-localised PM samplers of each type at both sites whether there is good repeatability in measures of oxidative potential form parallel TEOM and FDMS filters across the same period at the same site?

3 Methods

3.1 Monitoring Methods

The TEOM is a real time particulate mass monitor; its mass measurement method relies on the microbalance. This consists of a hollow glass tapered tube, clamped at one end free to oscillate at the other; an exchangeable Teflon coated glass fibre filter is placed on the free end. This tube is maintained in constant oscillation through an electronic feedback system, adding sufficient energy to the system to overcome losses. The frequency of oscillation is proportional to the mass and was measured and recorded by a microprocessor at two-second intervals. The TEOM was operated in its standard configuration. Air was drawn air through an R&P PM_{10} sampling inlet at 16.7 $lmin^{-1}$. The flow was then split using an isokinetic flow splitter into a main flow of 3 $lmin^{-1}$, which passed through the microbalance, and an auxiliary flow of 13.7 lmin-1. The filter and the air stream were heated to 50ºC to reduce the interferences from particle bound water and to minimize thermal expansion of the tapered element that may affect the oscillating frequency.

The FDMS measures the mass concentration of airborne PM and quantifies the mass changes of the filter due to evaporative and condensation processes. This system is based on TEOM technology, using the same microbalance. The FDMS sampled air through an R&P PM_{10} inlet, and then employed a dryer to remove water from the sample, this allowed the microbalance to be operated at 30ºC rather than 50ºC. After passing through the dryer, measurement was alternated between two modes (base and purge), switching between them every six minutes. The change in mass on the filter was measured by the microbalance during both modes. The base measurement was made by the microbalance after size selection and passing through the dryer. This provided a mass concentration of PM_{10}. The purge measurement was made after the sample stream had passed through the Pallflex membrane purge filter, chilled to 4 ºC, to remove particulate matter and low molecular weight organic compounds. This purged air was passed through the microbalance filter and the change in mass of filter measured. This provided in a mass concentration due to evaporative and condensation processes on the filter.

TEOMs PM_{10} instruments and FDMSs PM_{10} and $PM_{2.5}$ instruments were deployed at two sites (Birmingham and Teddington) as part of DEFRAs PM_{10} equivalency trial. Two of each type of instrument were operated simultaneously at each site as part of this trial.

Routine filter changes were scheduled on the same day every two weeks as both sites to ensure that sampling periods were approximately equal and that they sampled during broadly similar atmospheric conditions. The PM mass collected on both the FDMS and TEOM filters is calculated from the mass concentration calculated by the microprocessor, the known flow rate and the known exposure time.

3.2 Particle extractions and re-suspension

The established methods for PM extraction from Teflon filters used in the laboratory of Professor Kelly and Dr Mudway (2) were modified in the current study to ensure that volatile components were retained on the FDMS filters obtained at 4 and 30°C. All TEOM filters were collected directly into 50ml falcon tubes containing 5ml of chilled HPLC-grade methanol and immediately placed in a fridge for storage at 4°C. The FDMS filters had a larger diameter and were therefore stored in 20ml of methanol to ensure complete immersion of the filter. All filters were ultimately transported to King's College London for extraction and analysis in chilled iceboxes. Filter extraction was achieved by vortexing the filters in their methanol storage medium for 2 cycles of 5 minutes. Thereafter the filters underwent probe sonication (15 micron amplitude) for 30 seconds on ice. The filter substrate was then removed from the extraction tube and the methanol/particle extract dried down under nitrogen. The drying down procedure was performed at the temperature at which the filters were collected: 50°C for the standard TEOM PM_{10} filter, 30°C for the FDMS PM_{10} and $PM_{2.5}$ filters, and 4°C for the PM_{10} and $PM_{2.5}$ FDMS purge filters. The dried down particle extract was then re-suspended at a stock concentration of in Chelex100-resin treated (3g/100ml, mixed o/n at 4°C) ultra-pure (18Ω) containing 5% HPLC-grade methanol. The resin was removed by centrifugation (3,000 rpm for 15 minutes, 4°C) and the purified 5% methanol solution carefully decanted. Prior to use the solution had its pH adjusted to neutrality using Chelex resin treated 0.1M HCl or 0.1M KOH. Samples were re-suspended at the 150µg/ml by addition of a set volume of Chelex treated

water containing 5% methanol by vortexing for ten minutes followed by a further 30 second sonication as outlined above.

3.3 Particle incubations in the synthetic RTLF and antioxidant analysis

Particle suspensions corresponding to each filter were diluted to 55.56µg/mL in ultra-pure Chelex-resin treated water (pH7.4). To 225µl aliquots of each particle suspension 25µL of a composite stock antioxidant solution was added (2mM ascorbate, urate and reduced glutathione, pH7.4) to give a final particle concentration in each sample of 50µg/mL and antioxidant concentrations at time zero of 200µM. Samples were then transferred to a 37^{o}C incubator where they were gently mixed for a period of 4-hours. All incubations were performed in triplicate. At the end of this incubation period samples were acidified with MPA (final concentration 5%) to quench oxidation reactions, prior to transferring the samples to a centrifuge to spin out the particles (13,000 for 1-hour at 4^{o}C). The resultant supernatants were then processed for HPLC analysis of ascorbate and urate (reverse-phase with EC-detection, 0.5µAmps, 400mVolts (11)) and spectrophotometric determination of reduced glutathione and glutathione disulphide (12). All antioxidant concentrations after the four-hour incubation were compared against time 0 and time 4 control antioxidant values in particle free control samples.

3.4 Statistics

Comparison of antioxidant concentrations at the 4-hour time point was performed using a one-way ANOVA. Post hoc analyses of the groups were performed using paired t-tests reflecting the parallel nature of the collected samples. All analysis were performed treating the parallel filters measurements as separate samples. All association analyses were performed using the Pearson correlation. In all cases the null hypothesis was rejected at the 5% level. All statistical analyses were performed using the Unistat (Unistat, Ltd, London, UK) and SPSS (SPSS inc, Chicago, USA) packages on a Windows based PC platform.

4 Results

4.1 Particle collections

TEOM and FDMS filters were obtained from two co-localized samplers situated in Birmingham and Teddington over three separate sampling periods as indicted in Table 1Table. Only single FDMS $PM_{2.5}$ filters at 4 and 30°C were obtained for the first and second sampling period from Birmingham. Similarly only single FDMS PM_{10} filters at 4 and 30°C were obtained for the first sampling period at this site. Notably for the first collection period, ambient air was sampled at the Birmingham site between 30/11/04 – 22/12/04, compared with a longer interval, 15/11/04 – 22/12/04, at the Teddington site. This period difference is reflected by the substantially larger PM mass collected at the Teddington site over the first sampling period.

Table 2: Mass of TEOM and FDMS $PM_{2.5}$ and PM_{10} collections over the three sampling periods.

SITE	FILTER	COLLECTED PM MASS (mg)					
		15/11/04 – 22/12/04		22/12/04 – 20/01/05		20/01/05 – 17/02/05	
		A	B	A	B	A	B
Birmingham	FDMS $PM_{2.5}$ (4°C)	681.7	NS	314.3	NS	618.2	727.9
Birmingham	FDMS $PM_{2.5}$ (30°C)	681.7	NS	314.3	NS	628.2	727.9
Teddington	FDMS $PM_{2.5}$ (4°C)	1323.1	1283.3	383.6	395.7	781.4	770.7
Teddington	FDMS $PM_{2.5}$ (30°C)	1323.1	1283.3	383.6	395.7	781.4	770.7
Birmingham	FDMS PM_{10} (4°C)	NS	924.1	733.0	642.7	952.1	916.1
Birmingham	FDMS PM_{10} (30°C)	NS	924.1	733.0	642.7	952.1	916.1
Teddington	FDMS PM_{10} (4°C)	1744.0	1801.0	812.1	804.4	1120.0	663.0
Teddington	FDMS PM_{10} (30°C)	1744.0	1801.0	812.1	804.4	1120.0	663.0
Birmingham	TEOM PM_{10} (50°C)	1479.4	1448.7	1303.3	1318.8	1567.0	1587.1
Teddington	TEOM PM_{10} (50°C)	2915.4	2878.7	1496.4	1450.5	1794.5	1754.0

A and B indicate the filters were obtained from two separate instruments over the sampling periods. NS = no filter obtained for the indicated period

4.2 Ascorbic acid depletion

Incubation of the synthetic RTLF with particles extracted from each of the filter types at 50µg/mL resulted in a significant ($P<0.05$) loss of ascorbate over the 4-h incubation period relative to the 4-hour particle free control. These data are illustrated in Table 2Error! Reference source not found.. No loss of ascorbate was noted over the 4h incubation in the particle free control. Considering filters obtained from both sites across all three sampling periods we saw no difference between the extent of ascorbate depletion depending on whether PM were collected by standard TEOM at 50°C, or by FDMS at 4°C or 30°C (Table 2Error! Reference source not found.). When the data were separated into the Birmingham and Teddington collections (Figure 1) we observed a significantly ($P<0.05$) greater loss of ascorbate with both the $PM_{2.5}$ filters compared with the PM_{10} samples collected from the Birmingham site at both sampling temperatures. No difference was noted however between the extent of ascorbate depletion seen with either FDMS filter type at 4 and 30°C. The activity of the PM extracted from the TEOM filter collected at 50°C was significantly less than that seen with the PM_{10} filter collected at 30°C but not at 4°C, nor the $PM_{2.5}$ filters collected at 4 and 30°C. No differences were noted between the activities of PM extracted from any filter type from the Teddington site (Figure 1 – *lower panel*). It should be noted that whilst a full set of six paired filters were available from the Teddington site, due to instrument failures only 4 parallel filter collections were obtained from Birmingham (Table 1Table). Consequently the statistical differences

between PM_{10} activities seen between the TEOM at 50°C and the FDMA collected at 30°C should be interpreted with caution. No differences in the activities of PM extracted from any of the filter types over the three sampling intervals were noted between the two sites.

Table 3: Concentrations of antioxidants remaining in the synthetic RTLF following a 4h incubation with 50µg/ml PM samples derived from TEOM and FDMS filters collected from both Birmingham and Teddington.

Filter type	Ascorbate (mM)	Urate (mM)	Glutathione (mM)
Control Time 0	200.0±5.4 (n=3)	200.0±4.2 (n=3)	184.7±6.2 (n=3)
Control Time 4	197.3±5.6 (n=3)	202.5±3.6 (n=3)	155.9±4.2 (n=3)a
$PM_{2.5}$ – FDMS 4ºC	63.6±51.6* (n=10)	206.9±2.5 (n=10)	98.5±19.6* (n=10)
$PM_{2.5}$ – FDMS 30ºC	79.4±39.0*(n=10)	206.7±3.0 (n=10)	77.0±23.0* (n=10)
PM_{10} – FDMS 4ºC	78.1±49.9*(n=11)	206.9±3.0 (n=11)	87.8±30.2* (n=11)
PM_{10} – FDMS 30ºC	83.7±40.7* (n=11)	206.7±4.0 (n=11)	86.3±28.8* (n=11)
PM_{10} – TEOM 50ºC	74.6±43.9* (n=12)	206.2±3.2 (n=12)	84.5±20.5* (n=12)

All data are represented as means (standard deviation) of the indicated number of PM samples. These include PM extracted from both parallel filters at each site – indicated as A and B in the legend to Table 1. The asterisks indicates that the concentration of antioxidant remaining in the RTLF following incubation with the particle suspensions derived from each of the filter types was significantly different from that observed in the 4h particle-free control; 'a' indicates a significant loss of GSH from the particle free control over the 4h incubation period.

4.3 Reduced glutathione depletion

Data are illustrated in Table 1Table (all filters extracted PM from both sites) and Figure 2 (filters separated by site). We observed no difference between the oxidative activities of PM extracted from any of the filter types when both the Birmingham and Teddington sites were considered together (Table 1Table). When the data were separated between the two sites we observed that FDMS $PM_{2.5}$ samples obtained at 4°C were significantly more active than PM_{10} samples collected at 4°C. In addition FDMS PM_{10} samples collected at 30°C appeared to have greater activity than those collected at 4°C. Notably the 50°C TEOM PM collection did not appear to be significantly more active than either fraction of the FDMS collections at both collection temperatures. In contrast to the ascorbate data a small but significant loss of GSH was noted in the particle free controls over the 4h incubation period. No differences in the activities of PM extracted from any of the filter types over the three sampling intervals were noted between the two sites.

4.4 Urate depletion

We observed no losses of urate from the synthetic RTLF after with any of the PM samples derived from the various filter types. In addition urate appeared stable in the particle free control over the duration of the incubation period (Table 3Table **,** Figure 3).

4.5 Correlation analyses

The extent of ascorbate depletion from synthetic RTLF by parallel PM samples collected by TEOM at 50°C or FDMS at 4 and 30°C were strongly associated with each other with r values ranging from 0.72 – 0.91 (Figure 4). This contrasted with the lack of association seen between the FDMS collected PM (both $PM_{2.5}$ and PM_{10}) and that obtained using a standard TEOM with respect to their capacity to deplete glutathione depletion, r = 0.05 – 0.48 (all tested interactions non-significant – Figure 5). Notably whilst there was no association between TEOM and FDMS PM samples with respect to their ability to deplete glutathione, the separate FDMS collections at 4 and 30°C had broadly similar reactivity's: FDMS $PM_{2.5}$ at 4°C versus FDMS $PM_{2.5}$ at 30°C, r=0.80, P=0.005; FDMS PM_{10} at 4°C versus FDMS PM_{10} at 30°C, r=0.62, P=0.04 (Figure 5).

Strong correlations were observed between the extent of ascorbate and glutathione depletion seen with both the FDMS $PM_{2.5}$ and PM_{10} samples (Figure 6): for $PM_{2.5}$ r=0.74, P<0.001 and

for PM_{10} r=0.46, P=0.03. A similar though non-significant trend was also noted for the TEOM extracted PM samples: r=0.52, P=0.08.

As two separate TEOM and FDMS instruments were co-localised at each of the two sites it was possible to examine the repeatability of the measures of oxidative activity derived from PM extracted from parallel filters. A strong associated was noted for the determined oxidative activity, both in terms of ascorbate and glutathione depletion for PM extracted from the parallel TEOM filters: r=0.996, P<0.0001 and r=0.98, P<0.0001 respectively (Figure 7). Similar though weaker associations were noted for the FDMS $PM_{2.5}$ and PM_{10} samples (using data from both the 4^oC and 30^oC filters): FDMS $PM_{2.5}$ and PM_{10} ascorbate depletion, r=0.97, P<0.0001 and r=0.73, P=0.02 respectively; FDMS PM_{10} glutathione depletion, r=0.85, P=0.002. Of the FDMS samples only $PM_{2.5}$ glutathione depletion showed poor repeatability between the parallel filters (Figure 7). The strength of these underlying associations was not improved by considering only parallel filters collected at each of the sampling temperatures.

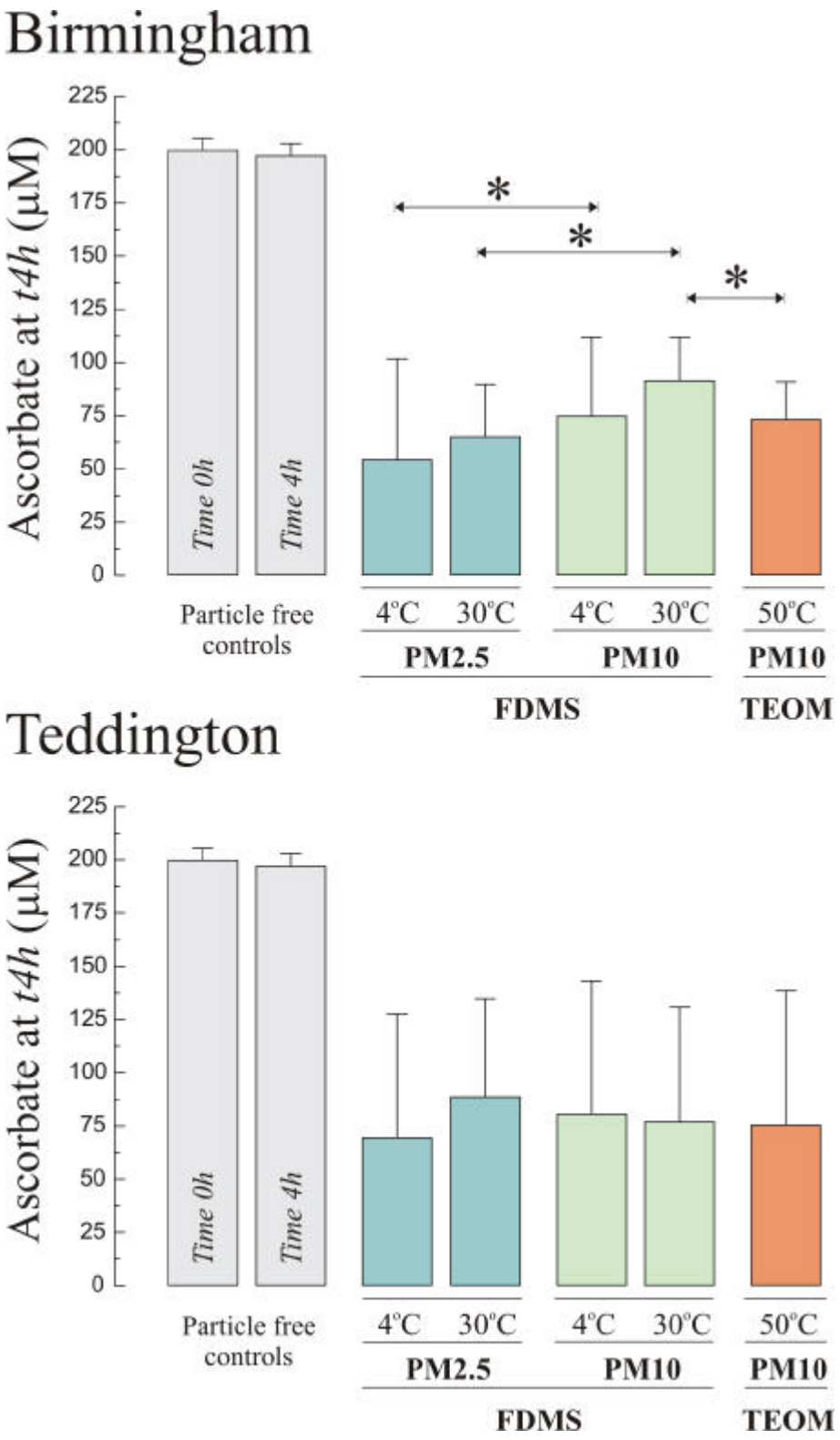

Figure 1: Concentration of ascorbate remaining in synthetic Respiratory Tract Lining Fluid) RTLF after a 4h incubation with 50mg/ml PM extracted from FDMS and TEOM filters collected under the indicated conditions.

All data are illustrated as means with standard deviation and 45 paired measurements for Birmingham and 6 paired samples for Teddington. An asterisk indicates that the means of the indicated groups differ significantly (P<0.05) based on a paired t-test.

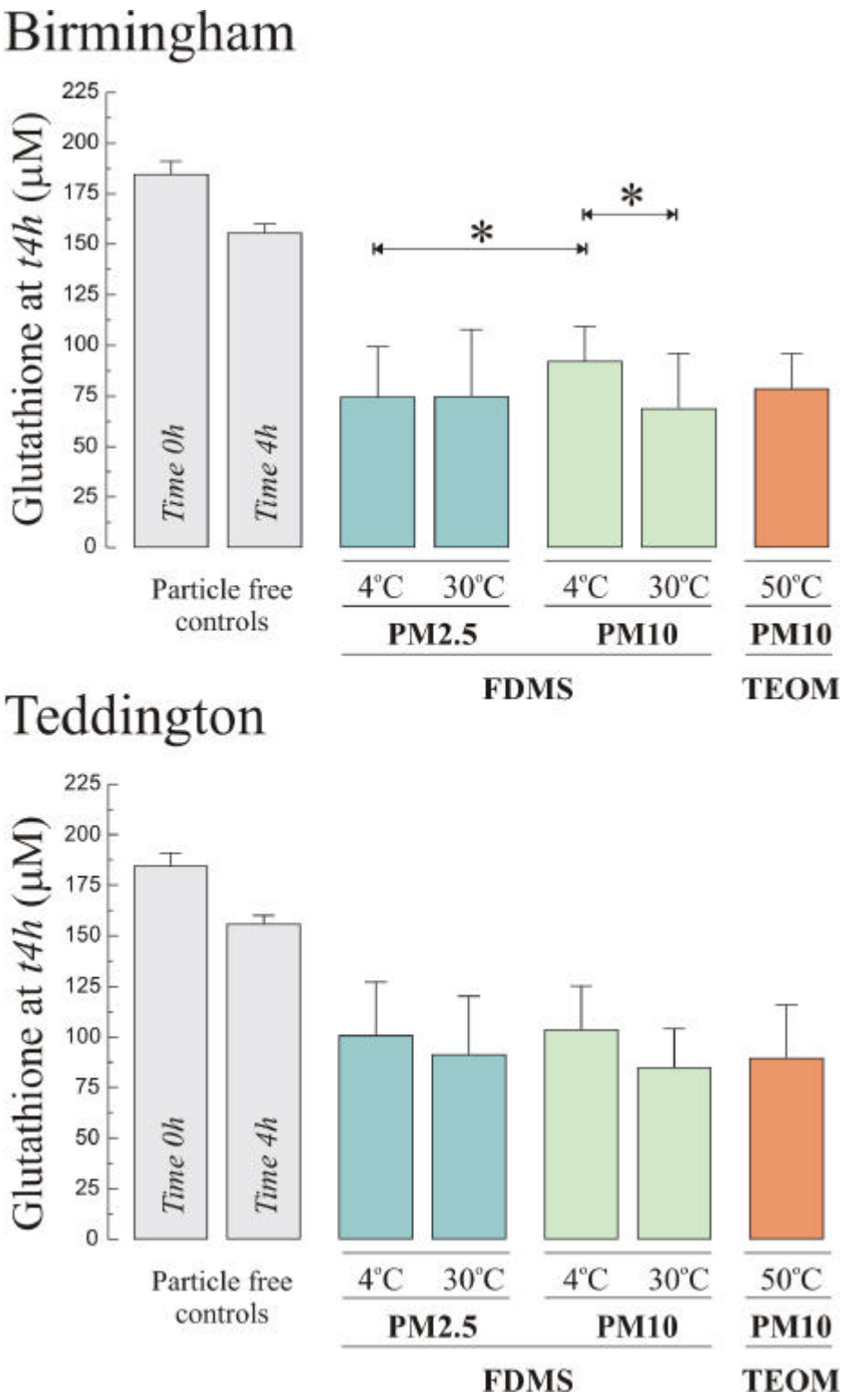

Figure 2:Concentration of glutathione remaining in synthetic RTLF after a 4h incubation with 50mg/ml PM extracted from FDMS and TEOM filters collected under the indicated conditions.

All data are illustrated as means with standard deviation and 45 paired measurements for Birmingham and 6 paired samples for Teddington. An asterisk indicates that the means of the indicated groups differ significantly (P<0.05) based on a paired t-test.

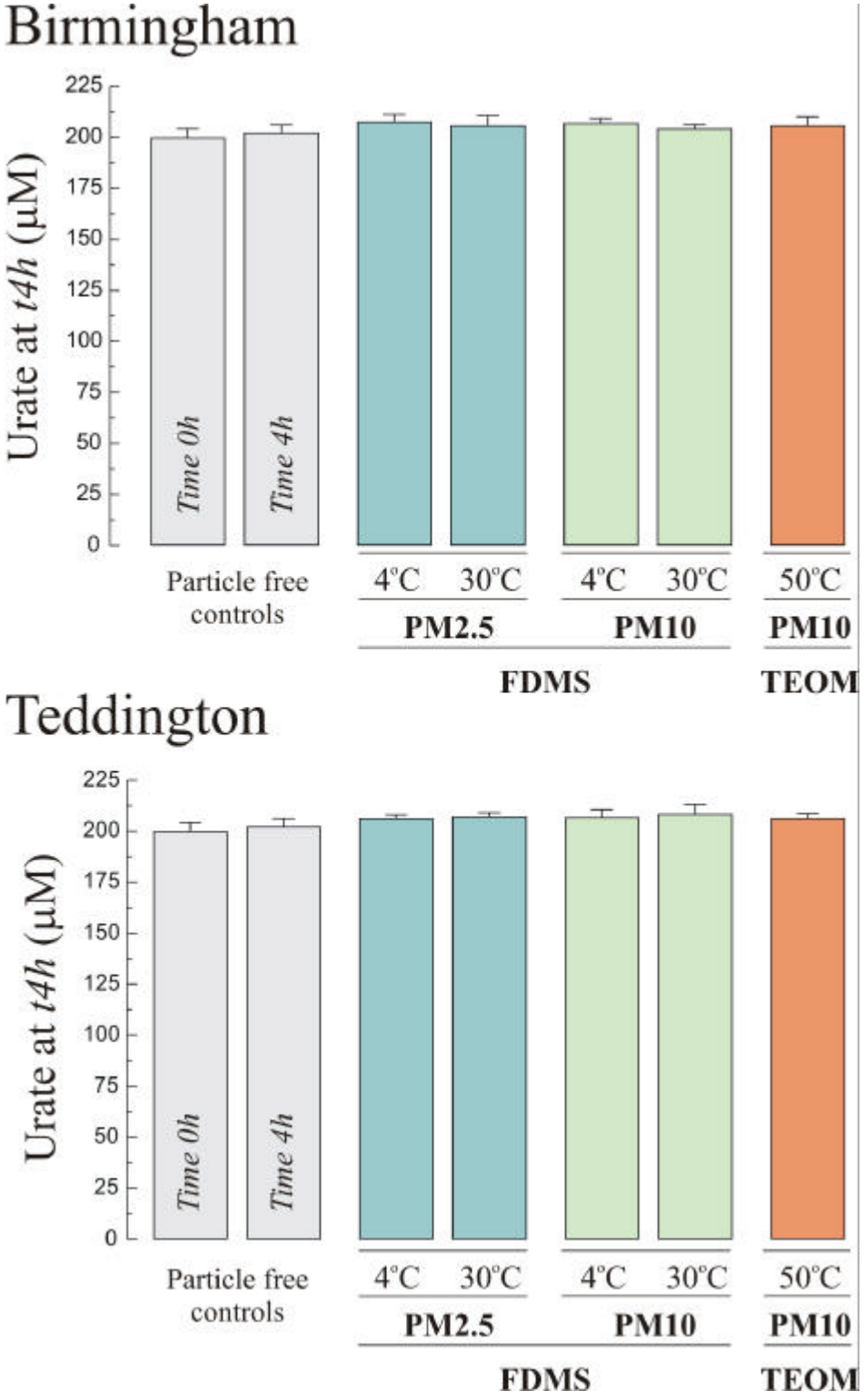

Figure 3:Concentration of urate remaining in synthetic RTLF after a 4h incubation with 50mg/ml Pm extracted from FDMS and TEOM filters collected under the indicated conditions.

All data are illustrated as means with standard deviation and 45 paired measurements for Birmingham and 6 paired samples for Teddington. An asterisks indicates that the means of the indicated groups differ significantly (P<0.05) based on a paired t-test.

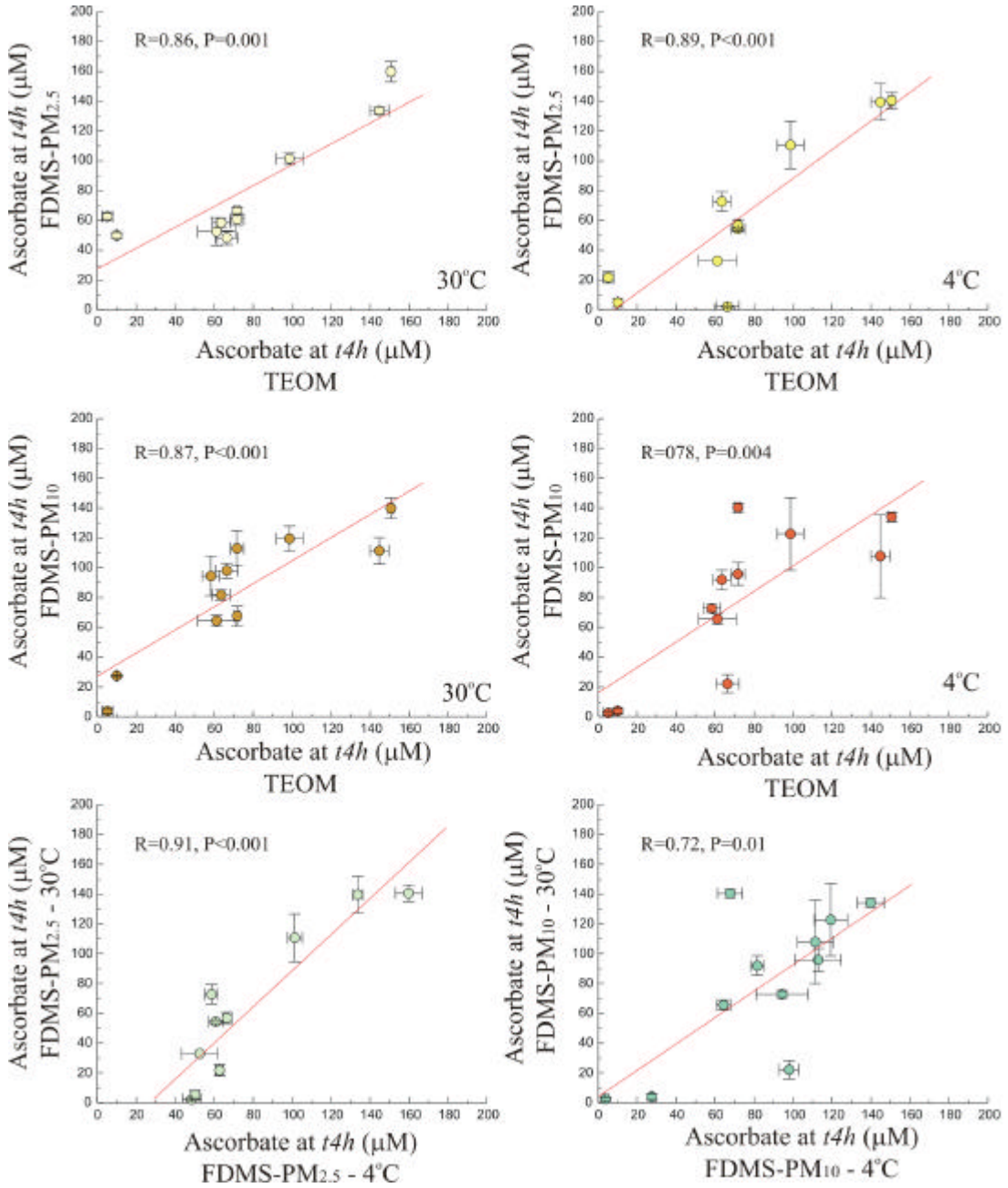

Figure 4: Association between the extents of ascorbate depletion from the synthetic RTLF by PM samples collected in parallel, either by standard TEOM at 50°C or by FDMS ($PM_{2.5}$ and PM_{10}) at 30 and 4°C.

Samples from both the Birmingham and Teddington sites are included for this correlative analysis. Each data point represents the mean (standard deviation) of triplicate determinations. The results of a Pearson's correlation analysis are inset for each set of comparisons along with a linear regression through the data to illustrate the degree of association.

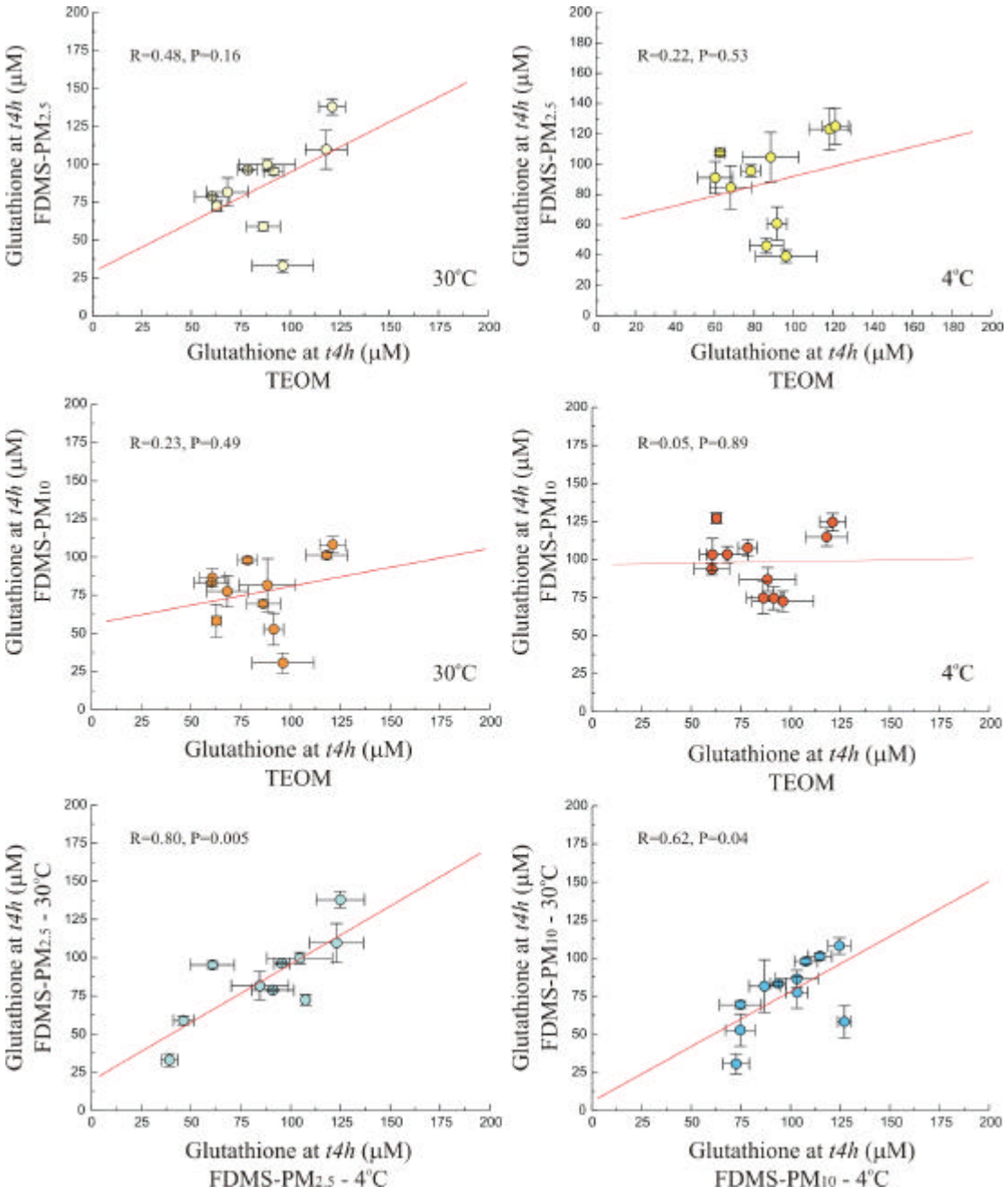

Figure 5: Association between the extents of glutathione depletion from the synthetic RTLF by PM samples collected in parallel, either by standard TEOM at 50°C or by FDMS ($PM_{2.5}$ and PM_{10}) at 30 and 4°C.

Samples from both the Birmingham and Teddington sites are included for this correlative analysis. Each data point represents the mean (standard deviation) of triplicate determinations. The results of a Pearson's correlation analysis are inset for each set of comparisons along with a linear regression through the data to illustrate the degree of association.

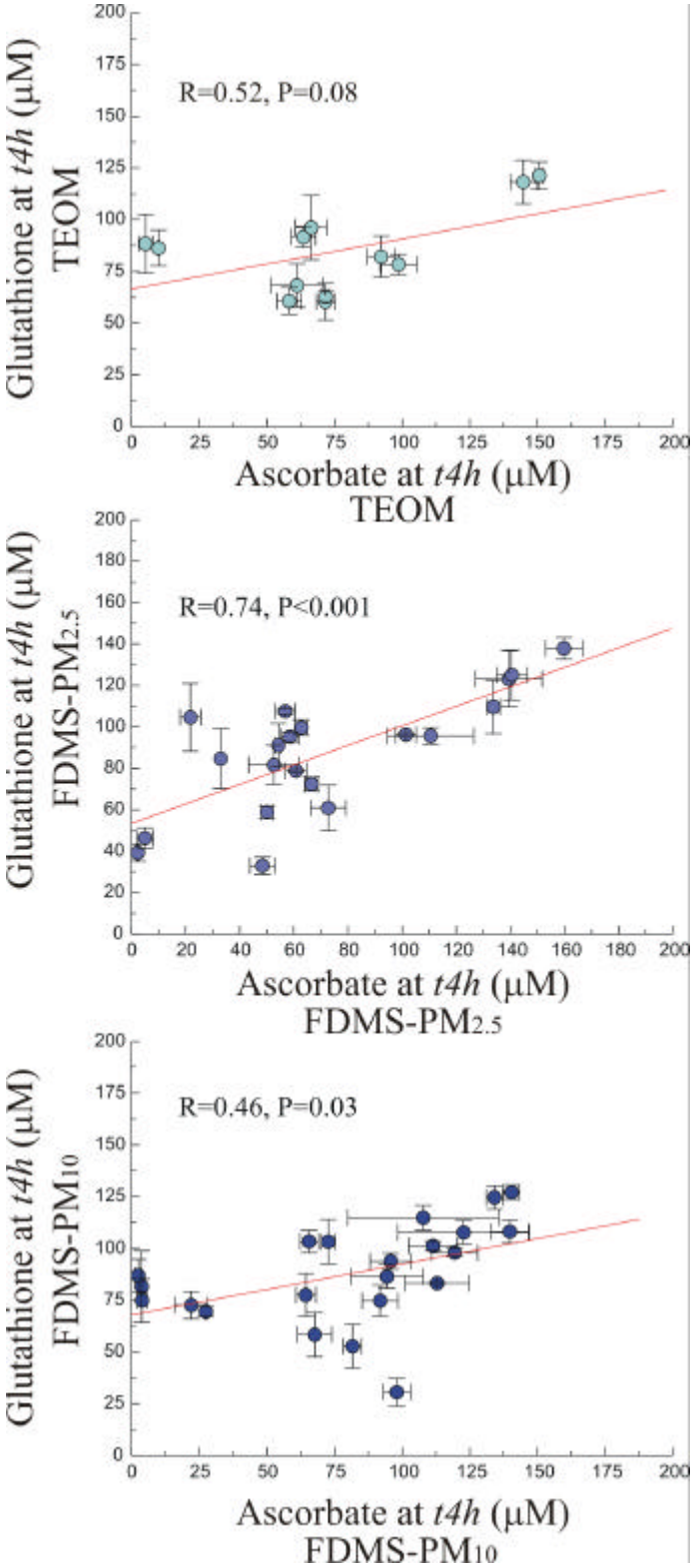

Figure 6:The degree of association between the extent of ascorbate and glutathione depletion seen by PM obtained either from the standard TEOM, operating at 50°C, or the FDMS ($PM_{2.5}$ and PM_{10}) operating 4 and 30°C.

Samples from both the Birmingham and Teddington sites are included for this correlative analysis. Each data point represents the mean (standard deviation) of triplicate determinations. The results of a Pearson's correlation analysis are inset for each set of comparisons along with a linear regression through the data to illustrate the degree of association.

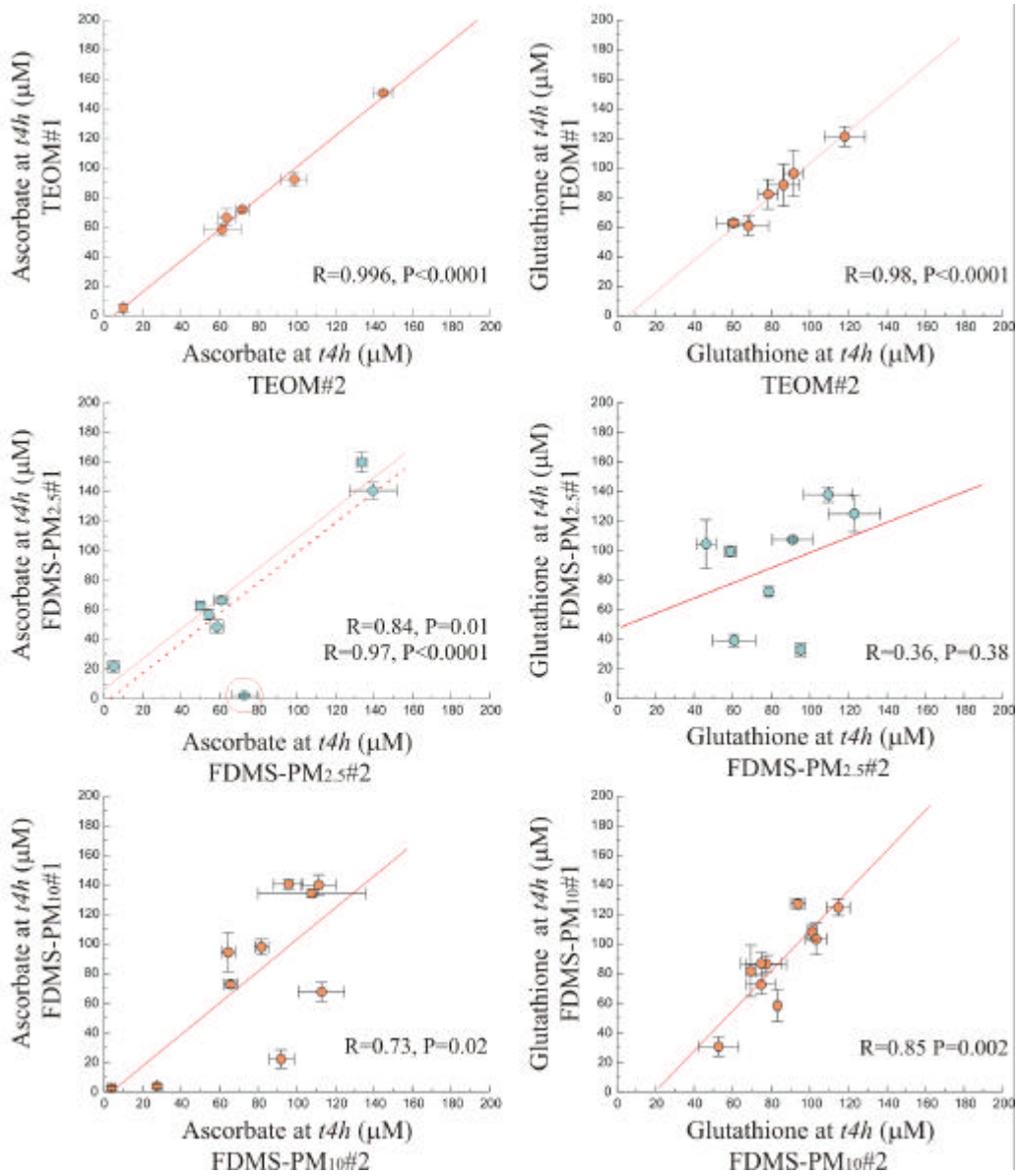

Figure 7: Repeatability of PM oxidative activity (ascorbate and glutathione) measurements determined using parallel TEOM and FDMS instruments at Birmingham and Teddington.

Samples from both the Birmingham and Teddington sites are included for this correlative analysis. For the FDMS comparisons the data obtained from the filters collected at both 4 and 30^{o}C are included in the correlation analysis.. Each data point represents the mean (standard deviation) of triplicate determinations. The results of a Pearson's correlation analysis are inset for each set of comparisons along with a linear regression through the data to illustrate the degree of association. In the panel examining the degree of association for ascorbate depletion between the FDMS $PM_{2.5}$ collections the correlation analysis results are given for the whole data, and after removing the outlying point, which has been circled.

5 Discussion

In the current study we investigated the hypothesis that volatile PM components not collected by the standard TEOM would contribute to the measured PM oxidative activity, and hence that PM extracted from standard TEOM filters would tend to underestimate the toxicity of ambient PM. To test this hypothesis we collected parallel PM samples using TEOM and FDMS instruments at two sites within the UK, Birmingham and Teddington. The oxidative activity of PM extracted from filters collected at 50, 30 and 4°C were then compared to establish whether TEOM sampling resulted in a loss of activity. All filters were maintained at 4°C after collection and the extraction procedures were performed at the same temperature at which the PM was collected to avoid post-sampling losses of volatile components.

The following questions were addressed in the current study:

- Is the toxicity of the sample collected at 4ºC greater than that of PM samples collected at 50oC (standard TEOM) consistent with the view that volatile PM components contribute significantly to PM oxidative activity?

We found no evidence to suggest that PM samples collected using the FDMS instrument at 4°C and 30°C were more active than samples obtained in parallel using the standard TEOM (50°C). Some minor differences were apparent in the Birmingham samples but these were quantitatively small and probably reflectthe limited number of filter comparisons that could be performed at this site. It was notable that whilst there was a good quantitative association between the TEOM and FDMS extracted PM samples with regard to their capacity to deplete ascorbate, there was rather poor agreement between the TEOM PM_{10} and the FDMS PM_{10} and $PM_{2.5}$ filter-extracted PM in terms of their ability to oxidise glutathione . This may indicate that although the overall activity was comparable the PM components contributing to the oxidative activity may have differed. For example certain PAHs and quinone species are known to form conjugates with GSH (13), whilst losses of AA are largely attributable to redox reactions with metal and quinine/hydroquinone species (5,6). Clarification of the basis for this poor agreement between glutathione and ascorbate depletion would require detailed chemical characterisation of the PM extracts obtained from the various filter types. This was not included in this contract.

- Whether there is a measurable difference in the oxidative potential of FDMS TEOM filter PM_{10} and $PM_{2.5}$ collected at 4 and 30°C?

As with the comparison to the standard TEOM no differences in the activity of the PM obtained at 4 or 30°C using the FDMS instrument were seen. Further, there was a strong quantitative relationship between the capacity of both individual paired $PM_{2.5}$ and PM_{10} filter extracted PM to deplete both ascorbate and reduced glutathione.

- Whether there is a site-specific difference in the oxidative potential between the two sites?

Whilst the oxidative activity (both in terms of ascorbate and glutathione depletion) of the PM samples obtained from both instruments and across all sampling temperature varied markedly at each site with time, there was no overall difference in the activity of PM between Birmingham and Teddington. This is unsurprising as the site locations are similar background sites, over 20 metres from major roads. The first in the campus of the Birmingham University, the second in the grounds of the National Physical Laboratory.

- As there are two co-localised PM samplers of each type at both sites whether there is good repeatability in measures of oxidative potential form parallel TEOM and FDMS filters across the same period at the same site?

The paired TEOM filter extracted PM samples obtained from co-localised instruments at both sites gave a remarkably high degree of agreement both in terms of ascorbate and glutathione depletion. The FDMS results were a little patchier but overall a good degree of agreement was seen between separate filters collected at the same site over comparable intervals. It should be noted that the extraction procedure that was used in these experiments has been optimised for TEOM microbalance filters and it may be that the procedure is not ideal for the extraction of

material from FDMS purge filters. This may underlie the greater variability that was apparent in the repeated FDMS measurements.

6 Conclusions

We found no evidence to support the view that volatile material lost as a consequence of the heating of the TEOM to 50^{o}C contributed to the measured oxidative activity of the sampled PM. In light of these findings we believe the use of PM extracted from standard TEOM filters will give an accurate estimate of the oxidative activity of the sampled PM air shed.

7 References

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