Seasonal survey of the composition and degradation state of particulate organic matter in the Rhone River using lipid tracers (written by M.-A. Galeron et al.)

General comments:

This paper provides detailed data on seasonal variation of a wide variety of lipid molecules contained in suspended particulate matter in the Rhone River estuary. Using these data, the authors discussed the origins and the biological and abiotic degradation processes of the lipids. In particular, their approach for evaluating quantitatively the influence of abiotic oxidation on the removal of a few lipid compounds seems an interesting challenge. Even though similar studies have been published for a few other ecosystems recently (e.g. Rontani et al. 2009, 2014b), I think that their study has a sufficient merit to be published in Biogeosciences after appropriate revision, because this study focused on an environment with relatively high human impacts compared to the preceding studies. The argument concerning the provenance of POM based on the lipid composition is generally convincing.

My concern is that their conclusions, particularly those for the individual degradation processes, depend on numerous assumptions that are not verified enough. Above all, the estimation of the degrees of biodegradation and auto- and photo-oxidation undergone by specific lipids such as cholesterol and sitosterol depends on the assumption that the parent lipids and all the degradation intermediates used for the estimation have similar turnover rates in river water, which seems dubious. In fact, the authors noted that some of the intermediates (e.g. Δ^6-5α-hydroperoxides, p.14213, line 5) are too unstable under certain conditions to be used as tracers. They used an alternative, apparently more stable species as a tracer; however, it seems quite difficult to confirm whether it is so stable as to conform to the above assumption. The assumption that the yield ratios of different oxidation intermediates are constant is also questionable. Although they mentioned these assumptions only briefly (p.14212, lines 2-5; p.14213, lines 1, 10-13), I would like to request them to elaborate the validity of these assumptions in more detail and discuss how the following interpretations may be changed if the turnover rates and/or the yield ratios are variable.

Specific comments:

- The use of the terms "plant-derived organic matter" and "organic matter of human-origin" in Abstract (lines 13-16) is an overgeneralization. This study investigated the degradation processes of only two sterols, one of which (sitosterol) is surely of plant origin but should

not be regarded as a representative organic matter of plants. The other, cholesterol, is not limited to human (*cf.* p.14206, lines 7-9).

- The authors mentioned that the sampling station at Arles was an estuarine station (p.14201, line 1). If so, the mixing between seawater and river water would play an important role in the dynamics of particulate matter. But they didn't argue this point in this paper. I would request them to show the salinity data in Fig. 2 and discuss briefly possible influence of salinity on the behavior of suspended particles. Water temperature data are also worthwhile to show here.
- Acidification with sulfuric acid (p.14201, line 13) is relatively rare for the treatment of POM samples. Can the authors refer to some reference paper? I guess excess sulfur may cause rapid deterioration of catalysts in the instrument.
- Measuring silicate using the GF/F filtrates (p.14201, lines 15-16) often leads to an overestimation due to leaching from the GF/F.
- A mixture of ethyl acetate/BSTFA (p.14202, line 24): what proportion?
- The combined DCM extracts (p.14203, line 4): Did it include also the chloroform phase of the initial phase separation, or not?
- The authors mentioned the minimum and maximal daily flow rates of the Rhone River recorded on 8 Oct 2011 and 19 May 2013, respectively (p.14204), but I could not find such sampling dates in Fig. 2. In addition, the time axis in Figs. 2-6 seems a bit confusing. Please consider adopting a real time scale where it is possible.
- "Typical for river systems" (p.14204, line 18): Literature should be referred to here.
- On page 14207 (lines 5-8), the authors suggested that 5 April 2011, 2 May 2013, and 4 Nov 2011 were flood dates. However, Fig. 2 shows that the river was under the base-flow conditions on 5 April 2011, and that the flow rate was not recorded on 4 Nov 2011.
- What do the authors mean by "content variability" (p.14208 line 5)?
- Section 3.2.4 (p.14208-9): The authors may explicitly mention here that the provenance analysis depending only on the fatty acid composition likely leads to an underestimation of higher plant contribution.
- I recommend that the definitions and calculation methods for CPPI (p.14211), biodegradation % (p.14212), and auto- and photo-oxidation % (p.14213) may be described in the Materials and Methods section.
- On page 14216 (lines 26-29), the authors mentioned the detection of *cis* and *trans* allylic 18,(8-11)-dihydroxyoleic acids (auto- and photo-oxidation intermediates) referring to

Table 2 and Fig. 5, but no such data can be found in this figure. Table 2 only shows 18,(8-11)-dihydroxy-$C_{16:0}$. They suggested a high proportion of *cis* isomers (line 29) and larger amounts of oxidation products than the parent ω-hydroxyoleic acid on a few sampling events (p.14217, lines 1-3), but these are not confirmed by the presented data.

- On page 14217 (lines 3-5), the authors mentioned "the previously discussed yearly variability in cuticular wax content", but it is a bit unclear what part of this manuscript they indicated by this phrase. Fig. 3b may be referred to here.
- What do the authors mean by "high compartmentalization effects" (p.14218, line 2)? Is it same as the protection by waxy materials from degradation suggested on p.14213 (lines 24-25)?

Technical corrections:

- Line 10 of page 14199: et -> and
- Line 28 of page 14205: A -> As (?)
- Line 16 of page 14214: please remove "a" from McCalley et al., 1981a.
- Cauwet et al. (1990) that appears in the reference list (p.14220) does not appear in the main text.
- Please remove "a" from Kolattukudy, P.E. 1980a (p.14222).