**Table 1:** Solid particle samples studied.

|  |  |  |  |
| --- | --- | --- | --- |
|  | **Sample** | **Grape variety** | **Clarification process** |
| Red wine | Red 1 | Merlot | Flash-release extraction, 2 h-enzymatic maceration (rapidase Extra Fruit (DSM), 2 g/hL), direct pneumatic pressing, enzymatic decantation (rapidase Clear (DSM), 2 g/hL) at 2°C. |
| Red 2 | Cabernet-Sauvignon | Thermo-vinification, centrifugation, |
| Rosé wine | Rosé 1 | Cabernet | Direct pneumatic pressing, enzymatic decantation (rapidase Clear (DSM), 2 g/hL) at 2°C. |
| Rosé 2 | Marselan | idem |
| White wine | White 1 | Chardonnay | idem |
| White 2 | Maccabeu | idem |
| White 3 | Viognier | Direct pneumatic pressing, enzymatic decantation (rapidase Clear (DSM), 2 g/hL) at 2°C, “gross” sludge harvest after 2 h of settling and “fine” sludge harvest after 22 h of settling |

**Table** **2**: Experimental design.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  |  | **Sterol content (mg/L)** | | | | |
| **Experiment** |  | **0 (negative control)** | **1.5** | **3.0** | **4.5** | **6.0** |
| Preliminary experiment | Synthetic β-sitosterol added (mg/1.1L) | none | 1.65 | 3.30 | 4.95 | 6.60 |

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  |  |  | **Type of grape solids** | | | | | | |
| **Experiment** |  | **Negative control** | **Positive control with synthetic β-sitosterol** | **Red 1** | **Red 2** | **Rosé 1** | **Rosé 2** | **White 1** | **White 2** | **White 3** |
|  | Dry matter of sludges added (mg/g) | none | none | 45.02 | 48.28 | 76.73 | 65.87 | 32.68 | 24.28 | 24.27 |
| 1st set | Mass added (g/1.1 L)  (reached turbidity (NTU)) | none | 0.0011  (1) | 5.564  (26) | 1.577  (40) | 2.375  (22) | 3.612  (21) | 4.701  (24) | 4.015  (15) | 4.378  (37) |
| 2nd set | Mass added (g/1.1 L) (reached turbidity (NTU)) | none  (1) | none | 4.248 (30) | 0.987 (30) | 1.591 (30) | 2.172 (30) | 3.452 (30) | 6.366 (30) | 2.720 (30) |

**Table 3:** Phytosterol composition (mg/g of dry matter) of solid particles / repartition (% w/w).

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Sample** | **Red 1** | **Red 2** | **Rosé 1** | **Rosé 2** | **White 1** | **White 2** | **White 3** | **Mean ± sd** |
| β-Sitosterol | 2.81 ± 0.02  /75.7 | 7.03 ± 0.54  /74.5 | 4.07 ± 0.56  /75.1 | 3.67 ± 0.39  /75.6 | 7.41 ± 0.90  /84.5 | 8.89 ± 0.00  /79.0 | 2.86 ± 0.32  /79.2 | 5.25 ± 2.29 |
| Campesterol | 0.25 ± 0.01  /6.7 | 0.76 ± 0.03  /8.1 | 0.34 ± 0.03  /6.3 | 0.37 ± 0.06  /7.6 | 0.48 ± 0.05  /5.5 | 0.69 ± 0.06  /6.1 | 0.21 ± 0.03  /5.8 | 0.44 ± 0.20 |
| Stigmasterol | 0.18 ± 0.01  /4.9 | 0.54 ± 0.07/5.7 | 0.24 ± 0.03/4.4 | 0.26 ± 0.12/5.3 | 0.37 ± 0.08/4.2 | 0.50 ± 0.01  /4.5 | 0.14 ± 0.02  /3.9 | 0.32 ± 0.14 |
| Sitostanol | 0.20 ± 0.00  /5.4 | 0.42 ± 0.01  /4.5 | 0.34 ± 0.04  /6.3 | 0.35 ± 0.18  /7.2 | 0.25 ± 0.04  /2.8 | 0.70 ± 0.28  /6.2 | 0.17 ± 0.01  /4.7 | 0.35 ± 0.17 |
| Non-identified sterols | 0.27 ± 0.01  /7.3 | 0.68 ± 0.01 /  7.2 | 0.43 ± 0.02  /7.9 | 0.21 ± 0.01  /4.3 | 0.26 ± 0.05  /3.0 | 0.47 ± 0.05 /4.2 | 0.23 ± 0.03  /6.4 | 0.36 ± 0.16 |
| **Total** | **3.71** ± **1.03**  **/100** | **9.43** ± **2.57**  **/100** | **5.42** ± **1.49**  **/100** | **4.86** ± **1.35**  **/100** | **8.77** ± **2.83**  **/100** | **11.25** ± **3.32**  **/100** | **3.61** ± **1.07**  **/100** | **6.72** ±  **2.83** |

**Table 4:** Fatty acid composition (mg/g of dry matter).

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Name** | **Formula** | **Red 1** | **Red 2** | **Rosé 1** | **Rosé 2** | **White 1** | **White 2** | **White 3** | **Mean ± sd** |
| Palmitic | C16:0 | 5.35 ± 0.32 | 14.65 ± 1.41 | 6.10 ± 0.08 | 6.31 ± 0.30 | 11.69 ± 0.34 | 11.62 ± 2.12 | 4.52 ± 0.17 | 8.61 ± 3.96 |
| Palmitoleic | C16:1 | 0.43 ± 0.03 | 1.03 ± 0.07 | 0.30 ± 0.02 | 0.19 ± 0.02 | 0.61 ± 0.01 | 0.56 ± 0.09 | 0.17 ± 0.01 | 0.47 ± 0.30 |
| Stearic | C18:0 | 0.98 ± 0.05 | 2.67 ± 0.16 | 0.99 ± 0.10 | 0.87 ± 0.04 | 1.86 ± 0.10 | 1.74 ± 0.23 | 0.76 ± 0.04 | 1.41 ± 0.70 |
| Oleic, linoleic and linolenic | C18:1, C18:2, C18:3 | 10.86 ± 0.49 | 24.76 ± 1.68 | 10.64 ± 0.10 | 12.13 ± 0.53 | 22.00 ± 0.60 | 20.24 ± 4.03 | 7.74 ± 0.42 | 15.48 ± 6.67 |
| Arachidic | C20:0 | 0.13 ± 0.00 | 0.27 ± 0.04 | 0.09 ± 0.00 | 0.15 ± 0.01 | 0.20 ± 0.02 | 0.10 ± 0.01 | 0.04 ± 0.00 | 0.14 ± 0.08 |
| Heneicosylic | C21:0 | 0.48 ± 0.02 | 1.19 ± 0.05 | 0.42 ± 0.03 | 0.45 ± 0.02 | 1.21 ± 0.08 | 0.81 ± 0.12 | 0.30 ± 0.02 | 0.69 ± 0.38 |
| Behenic | C22:0 | 0.29 ± 0.02 | 0.78 ± 0.03 | 0.36 ± 0.02 | 0.44 ± 0.01 | 0.85 ± 0.07 | 0.54 ± 0.04 | 0.25 ± 0.01 | 0.50 ± 0.23 |
| Ticosylic | C23:0 | 1.04 ± 0.04 | 2.66 ± 0.13 | 1.11 ± 0.15 | 1.27 ± 0.07 | 2.25 ± 0.16 | 1.67 ± 0.23 | 0.78 ± 0.04 | 1.54 ± 0.62 |
| Lignoceric | C24:0 | 0.21 ± 0.01 | 0.52 ± 0.02 | 0.27 ± 0.04 | 0.22 ± 0.01 | 0.57 ± 0.05 | 0.45 ± 0.07 | 0.18 ± 0.01 | 0.35 ± 0.16 |
| Pentacosylic | C25:0 | 0.51 ± 0.02 | 1.29 ± 0.04 | 0.72 ± 0.08 | 0.82 ± 0.05 | 1.17 ± 0.10 | 0.71 ± 0.10 | 0.43 ± 0.02 | 0.81 ± 0.32 |
| Cetoric | C26:0 | 0.10 ± 0.00 | 0.26 ± 0.01 | 0.14 ± 0.00 | 0.15 ± 0.01 | 0.26 ± 0.02 | 0.18 ± 0.03 | 0.08 ± 0.01 | 0.17 ± 0.07 |
| Montanic | C28:0 | 0.57 ± 0.02 | 1.52 ± 0.59 | 0.93 ± 0.02 | 1.54 ± 0.10 | 1.64 ± 0.01 | 0.69 ± 0.03 | 0.50 ± 0.01 | 1.06 ± 0.50 |
| Melissic | C30:0 | 0.73 ± 0.03 | 0.75 ± 0.44 | 0.66 ± 0.03 | 1.10 ± 0.10 | 1.47 ± 0.06 | 0.82 ± 0.10 | 0.37 ± 0.01 | 0.84 ± 0.35 |
| Lacceroic | C32:0 | 0.41 ± 0.07 | 0.43 ± 0.01 | 0.33 ± 0.01 | 0.50 ± 0.01 | 0.51 ± 0.14 | 0.54 ± 0.05 | 0.12 ± 0.01 | 0.41 ± 0.14 |
| Geddic | C34:0 | 0.33 ± 0.01 | 0.42 ± 0.00 | 0.41 ± 0.02 | 0.43 ± 0.23 | 0.18 ± 0.08 | 0.55 ± 0.05 | 0.04 ± 0.00 | 0.34 ± 0.17 |
| **Total** |  | **22.42** ± **2.79** | **53.20** ± **6.64** | **23.47** ± **2.81** | **26.57** ± **3.14** | **46.47** ± **5.74** | **41.22** ± **5.42** | **16.28** ± **2.14** | **32.81** ± 12.99 |
| Unsaturated acids/total acids (%) |  | 50.35 | 48.47 | 46.61 | 46.37 | 48.66 | 50.45 | 48.59 | 48.50 ± 1.48 |

**Table 5:** Grape solid overall lipid composition content (mg/g of dry matter) / repartition (% w/w).

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Sample** | **Red 1** | **Red 2** | **Rosé 1** | **Rosé 2** | **White 1** | **White 2** | **White 3** | **Mean ± sd** |
| Total fatty acids | 22 ± 3  /85 | 53 ± 7  /84 | 23 ± 3  /82 | 27 ± 3  /84 | 46 ± 6  /85 | 41 ± 5  /79 | 16 ± 2  /80 | 33 ± 13 |
| Total phytosterols | 4 ± 1  /15 | 9 ± 3  /16 | 5 ± 1  /18 | 5 ± 1  /16 | 9 ± 3  / 15 | 11 ± 3  /21 | 4 ± 1  /20 | 7 ± 3 |
| Total lipids | 26  /100 | 63  /100 | 28  /100 | 32  /100 | 54  /100 | 52  /100 | 20  /100 | 39 ± 15 |

**Table 6**: Assimilated N at 80% of released CO2, residual sugars at the end of the fermentation and maximum yeast population reached during the first set of experiments.

|  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | **Negative control** | **Positive control** | **Red 1** | **Red 2** | **Rosé 1** | **Rosé 2** | **White 1** | **White 2** | **White 3** | **Mean ± sd**  **on musts with solids (n=7)** |
| Assimilated N (mg/L) | 227 ± 1 | 287 ± 4 | 261 ± 11 | 251 ± 15 | 255 ± 17 | 268 ±11 | 262 ± 6 | 273 ± 9 | 276 ± 12 | 264 ± 9 |
| Assimilated N (%) | 53 ± 0 | 68 ± 1 | 63 ± 3 | 59 ± 11 | 60 ± 5 | 63 ± 2 | 62 ± 1 | 64 ± 2 | 65 ± 3 | 62 ± 2 |
| Residual sugars (g/L) | 17 ± 0 | 21 ± 0 | 20 ± 0 | 17 ± 0 | 20 ± 0 | 19 ± 0 | 25 ± 0 | 10 ± 0 | 25 ± 0 | 16 ± 6 |
| Maximum yeast population  ( 106 cells/mL) | 51 ± 3 | 84 ± 7 | 77 ± 1 | 82 ± 8 | 83 ± 8 | 84 ± 8 | 79 ± 7 | 83 ± 8 | 84 ± 4 | 82 ± 3 |