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**Supplemental Table S1.** Multivariable generalized linear mixed model showing the effect of treatment, period within the treatment, lactation number, stage of lactation, and interaction of treatment and stage of lactation on machine milking-induced short term changes

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment4 |  | <0.0001 |  |
| TRT | −0.1 (0.1) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.4 (0.1)b |  | 1.07 (0.92−1.24) |
| Period 4 (TRT) | Referentc |  | 0.71 (0.60−0.84) |
| Period 1 (CON) | 0.4 (0.1)a |  | 1.52 (1.32−1.75) |
| Period 3 (CON) | Referentb |  | − |
| Lactation number |  | <0.0001 |  |
| 1st | 0.02 (0.05)a |  | 1.02 (0.93–1.12) |
| 2nd | −0.2 (0.05)b |  | 0.79 (0.71–0.88) |
| ≥ 3rd | Referenta |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 0.5 (0.1)a |  | – |
| 101–200 DIM | −0.02 (0.1)c |  | – |
| > 200 DIM | Referentb |  | – |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.4 (0.1)b |  | 1.00 (0.82–1.22) |
| CON × ≤ 100 DIM | Referenta |  | 1.60 (1.33–1.93) |
| TRT × 101–200 DIM | −0.4 (0.1)c |  | 0.60 (0.48–0.75) |
| CON × 101–200 DIM | Referentb |  | 0.98 (0.80–1.20) |
| TRT × > 200 DIM | Referentb |  | 0.93 (0.77–1.14) |
| CON × > 200 DIM | Referentb |  | – |

a–cGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc test.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S2.** Multivariable generalized linear mixed model showing the effect of treatment, period within the treatment, and lactation number on machine milking-induced teat-end hyperkeratosis

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment4 |  | 0.23 |  |
| TRT | 0.2 (0.1) |  | 1.05 (0.97−1.15) |
| CON | Referent |  | − |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | −0.01(0.06)a |  | 1.22 (1.04−1.43) |
| Period 4 (TRT) | Referenta |  | 1.23 (1.05−1.45) |
| Period 1 (CON) | 0.3 (0.1)a |  | 1.35 (1.14−1.59) |
| Period 3 (CON) | Referentb |  | − |
| Lactation number |  | <0.0001 |  |
| 1st | −1.3 (0.1)c |  | 0.28 (0.25–0.31) |
| 2nd | −0.6 (0.1)b |  | 0.58 (0.52–0.64) |
| ≥ 3rd | Referenta |  | – |

a–cGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc test.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S3.** Multivariable general linear mixed model showing the effect of treatment, period within the treatment, presence of non-lactating quarter, lactation number, stage of lactation, log10-transformed somatic cell count (logSCC), milking session, interaction of treatment with lactation number, and interaction of treatment with stage of lactation on milk yield (kg/session)

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | LSM (95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.2 (0.04) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.4 (0.03)b |  | 13.7 (13.6−13.8) |
| Period 4 (TRT) | Referentc |  | 13.2 (13.1−13.3) |
| Period 1 (CON) | 1.1 (0.02)a |  | 14.2 (14.1−14.3) |
| Period 3 (CON) | Referentd |  | 13.1 (13.0−13.2) |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | 1.4 (0.1) |  | 14.2 (14.2–14.3) |
| Present | Referent |  | 12.8 (12.6–13.0) |
| Lactation number |  | <0.0001 |  |
| 1st | −2.4 (0.1)c |  | – |
| 2nd | −0.4 (0.1)b |  | – |
| ≥ 3rd | Referenta |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 3.7 (0.1)a |  | – |
| 101–200 DIM | 2.5 (0.1)b |  | – |
| > 200 DIM | Referentc |  | – |
| LogSCC | −0.9 (0.1) | <0.0001 | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.4 (0.01)b |  | 13.4 (13.4–13.5) |
| Session 2 | 1.1 (0.01)a |  | 14.1 (14.0–14.2) |
| Session 3 | Referentc |  | 13.0 (13.0–13.1) |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | 0.2 (0.04)d |  | 12.0 (11.9–12.2) |
| CON × 1st | Referentd |  | 12.2 (12.0–12.3) |
| TRT × 2nd | 0.08 (0.04)c |  | 14.0 (13.8–14.1) |
| CON × 2nd | Referentb |  | 14.2 (14.0–14.3) |
| TRT × 3rd | Referentb |  | 14.3 (14.1–14.4) |
| CON × 3rd | Referenta |  | 14.5 (14.4–14.7) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | 0.03 (0.04)a |  | 15.2 (15.0–15.4) |
| CON × ≤ 100 DIM | Referenta |  | 15.3 (15.1–15.4) |
| TRT × 101–200 DIM | −0.3 (0.04)c |  | 13.7 (13.5–13.8) |
| CON × 101–200 DIM | Referentb |  | 14.1 (13.9–14.2) |
| TRT × > 200 DIM | Referentd |  | 11.5 (11.3–11.6) |
| CON × > 200 DIM | Referentd |  | 11.5 (11.4–11.7) |

a–dGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S4.** Multivariable general linear mixed model showing the effect of treatment, period within the treatment, lactation number, stage of lactation, milking session, log10-transformed somatic cell count (logSCC), interaction of treatment and lactation number, and interaction of treatment and stage of lactation on milking unit-on time (s)

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | LSM (95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.01 (0.001) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.02 (0.001)a |  | 240 (238−241) |
| Period 4 (TRT) | Referentc |  | 231 (230−232) |
| Period 1 (CON) | 0.02 (0.001)b |  | 236 (235−237) |
| Period 3 (CON) | Referentd |  | 225 (224−227) |
| Lactation number |  | <0.0001 |  |
| 1st | −0.03 (0.002)b |  | – |
| 2nd | ­­−0.01 (0.002)a |  | – |
| ≥ 3rd | Referenta |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 0.06 (0.003)a |  | – |
| 101–200 DIM | 0.03 (0.03)b |  | – |
| > 200 DIM | Referentc |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.01 (0.002)b |  | 234 (232–235) |
| Session 2 | 0.02 (0.002)a |  | 237 (236–238) |
| Session 3 | Referentc |  | 229 (227–230) |
| LogSCC | −0.02 (0.002) | <0.0001 | – |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | −0.002 (0.001)c |  | 224 (222–225) |
| CON × 1st | Referentd |  | 220 (218–222) |
| TRT × 2nd | 0.002 (0.001)a |  | 240 (238–242) |
| CON × 2nd | Referentb |  | 234 (232–236) |
| TRT × 3rd | Referenta |  | 242(241–245) |
| CON × 3rd | Referentab |  | 238 (236–240) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.004 (0.001)a |  | 251 (248–253) |
| CON × ≤ 100 DIM | Referentb |  | 247 (245–249) |
| TRT × 101–200 DIM | −0.004 (0.001)c |  | 236 (234–238) |
| CON × 101–200 DIM | Referentd |  | 232 (230–234) |
| TRT × > 200 DIM | Referente |  | 220 (218–222) |
| CON × > 200 DIM | Referentf |  | 214 (213–216) |

a–fGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S5.** Multivariable general linear mixed model showing the effect of treatment, period within the treatment, presence of non-lactating quarter, lactation number, stage of lactation, milking session, interaction of treatment and lactation number, and interaction of treatment and stage of lactation on 2-minute milk yield (kg)

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | LSM (95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | −0.4 (0.02) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | −0.1 (0.01)d |  | 6.7 (6.6−6.8) |
| Period 4 (TRT) | Referentc |  | 6.8 (6.7−6.9) |
| Period 1 (CON) | 0.05 (0.01)a |  | 7.2 (7.1−7.3) |
| Period 3 (CON) | Referentb |  | 7.1 (7.1−7.2) |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | 0.8 (0.1) |  | 7.3 (7.3–7.4) |
| Present | Referent |  | 6.6 (6.4–6.7) |
| Lactation number |  | <0.0001 |  |
| 1st | −0.9 (0.1)b |  | – |
| 2nd | −0.2 (0.1)a |  | – |
| ≥ 3rd | Referenta |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 0.6 (0.1)a |  | – |
| 101–200 DIM | 0.6 (0.1)b |  | – |
| > 200 DIM | Referentc |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | −0.1 (0.01)c |  | 6.8 (6.7–6.8) |
| Session 2 | 0.3 (0.01)a |  | 7.2 (7.1–7.3) |
| Session 3 | Referentb |  | 6.9 (6.8–7.0) |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | 0.3 (0.02)d |  | 6.4 (6.3–6.5) |
| CON × 1st | Referentc |  | 6.6 (6.5–6.7) |
| TRT × 2nd | 0.04 (0.02)b |  | 6.9 (6.7–7.0) |
| CON × 2nd | Referenta |  | 7.4 (7.2–7.5) |
| TRT × 3rd | Referentb |  | 7.0 (6.9–7.1) |
| CON × 3rd | Referenta |  | 7.5 (7.4–7.6) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | 0.1 (0.02)b |  | 7.1 (6.9–7.2) |
| CON × ≤ 100 DIM | Referenta |  | 7.4 (7.2–7.5) |
| TRT × 101–200 DIM | −0.2 (0.02)c |  | 6.7 (6.6–6.8) |
| CON × 101–200 DIM | Referenta |  | 7.3 (7.2–7.4) |
| TRT × > 200 DIM | Referentd |  | 6.4 (6.3–6.5) |
| CON × > 200 DIM | Referentc |  | 6.8 (6.7–6.9) |

a–dGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S6.** Multivariable general linear mixed model showing the effect of treatment, period within the treatment, presence of non-lactating quarter, lactation number, stage of lactation, milking session, interaction of treatment and lactation number, and interaction of treatment and stage of lactation on peak milk flow rate (kg/min)

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | LSM (95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.1 (0.01) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.001 (0.001) |  | 5.7 (5.6−5.7) |
| Period 4 (TRT) | Referent |  | 5.7 (5.6−5.7) |
| Period 1 (CON) | 0.01 (0.01) |  | 5.7 (5.6−5.7) |
| Period 3 (CON) | Referentc |  | 5.6 (5.5−5.6) |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | 0.5 (0.04) |  | 5.9 (5.9–5.9) |
| Present | Referent |  | 5.4 (5.3–5.5) |
| Lactation number |  | <0.0001 |  |
| 1st | −0.3 (0.04)c |  | – |
| 2nd | 0.2 (0.04)a |  | – |
| ≥ 3rd | Referentb |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 0.2 (0.04)a |  | – |
| 101–200 DIM | 0.3 (0.04)a |  | – |
| > 200 DIM | Referentb |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.02 (0.001)c |  | 5.6 (5.6–5.7) |
| Session 2 | 0.07 (0.001)a |  | 5.7 (5.7–5.7) |
| Session 3 | Referentb |  | 5.6 (5.6–5.7) |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | 0.05 (0.01)d |  | 5.4 (5.4–5.5) |
| CON × 1st | Referente |  | 5.3 (5.3–5.4) |
| TRT × 2nd | −0.01 (0.01)a |  | 5.9 (5.8–6.0) |
| CON × 2nd | Referentb |  | 5.8 (5.8–5.9) |
| TRT × 3rd | Referentb |  | 5.7 (5.7–5.8) |
| CON × 3rd | Referentc |  | 5.7 (5.6–5.7) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | 0.02 (0.01)bc |  | 5.7 (5.7–5.8) |
| CON × ≤ 100 DIM | Referentad |  | 5.7 (5.6–5.7) |
| TRT × 101–200 DIM | −0.06 (0.01)ab |  | 5.8 (5.7–5.8) |
| CON × 101–200 DIM | Referentcd |  | 5.7 (5.7–5.8) |
| TRT × > 200 DIM | Referente |  | 5.5 (5.5–5.6) |
| CON × > 200 DIM | Referentf |  | 5.5 (5.4–5.5) |

a–fGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S7.** Multivariable general linear mixed model showing the effect of treatment, period within the treatment, presence of non-lactating quarter, lactation number, stage of lactation, milking session, interaction of treatment and lactation number, and interaction of treatment and stage of lactation on duration of low milk flow rate (s)

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | LSM (95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.2 (0.003) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.02 (0.002) |  | 22 (21−22) |
| Period 4 (TRT) | Referent |  | 21 (20−21) |
| Period 1 (CON) | 0.002 (0.002) |  | 14 (14−15) |
| Period 3 (CON) | Referentc |  | 14 (14−15) |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | −0.04 (0.01) |  | 17 (16–17) |
| Present | Referent |  | 18 (18–19) |
| Lactation number |  | <0.0001 |  |
| 1st | 0.1 (0.01)a |  | – |
| 2nd | 0.1 (0.01)b |  | – |
| ≥ 3rd | Referentc |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | −0.05 (0.01)c |  | – |
| 101–200 DIM | −0.04 (0.01)b |  | – |
| > 200 DIM | Referenta |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.01 (0.001)a |  | 18 (18–19) |
| Session 2 | −0.05 (0.001)c |  | 16 (16–16) |
| Session 3 | Referentb |  | 18 (18–18) |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | −0.04 (0.003)a |  | 24 (23–24) |
| CON × 1st | Referentd |  | 17 (17–18) |
| TRT × 2nd | −0.01 (0.003)b |  | 21 (21–22) |
| CON × 2nd | Referente |  | 14 (14–15) |
| TRT × 3rd | Referentc |  | 19 (18–19) |
| CON × 3rd | Referentf |  | 12 (12–13) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.02 (0.003)c |  | 19 (19–20) |
| CON × ≤ 100 DIM | Referentf |  | 14 (13–14) |
| TRT × 101–200 DIM | 0.02 (0.003)b |  | 22 (21–22) |
| CON × 101–200 DIM | Referente |  | 14 (14–14) |
| TRT × > 200 DIM | Referenta |  | 23 (22–23) |
| CON × > 200 DIM | Referentd |  | 15 (15–16) |

a–fGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S8.** Multivariable generalized linear mixed model showing the effect of treatment, period within the treatment, non-lactating quarter, lactation number, stage of lactation, milking session, log10-transformed somatic cell count (LogSCC), interaction of treatment and lactation number, and interaction of treatment and stage of lactation on bimodality

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | −1.2 (0.01) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | −0.05 (0.01)c |  | 0.36 (0.35−0.37) |
| Period 4 (TRT) | Referentb |  | 0.38 (0.37−0.39) |
| Period 1 (CON) | −0.01 (0.01)a |  | 1.00 (0.98−1.01) |
| Period 3 (CON) | Referenta |  | − |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | −0.1 (0.01) |  | 0.89 (0.87–0.90) |
| Present | Referent |  | – |
| Lactation number |  | <0.0001 |  |
| 1st | 0.1 (0.01)b |  | – |
| 2nd | 0.2 (0.01)a |  | – |
| ≥ 3rd | Referentc |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | −0.7 (0.01)c |  | – |
| 101–200 DIM | −0.5 (0.01)b |  | – |
| > 200 DIM | Referenta |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.2 (0.01)a |  | 1.19 (1.17–1.20) |
| Session 2 | −0.2 (0.01)c |  | 0.82 (0.81–0.83) |
| Session 3 | Referentb |  | – |
| LogSCC | 0.1 (0.01) | <0.0001 | 1.08 (1.06−1.09) |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | 0.2 (0.01)e |  | 0.42 (0.41–0.43) |
| CON × 1st | Referentb |  | 1.07 (1.04–1.09) |
| TRT × 2nd | 0.1 (0.01)d |  | 0.45 (0.44–0.46) |
| CON × 2nd | Referenta |  | 1.19 (1.16–1.22) |
| TRT × 3rd | Referentf |  | 0.34 (0.33–0.35) |
| CON × 3rd | Referentc |  | – |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | 0.1 (0.01)f |  | 0.18 (0.17–0.18) |
| CON × ≤ 100 DIM | Referentc |  | 0.49 (0.48–0.50) |
| TRT × 101–200 DIM | 0.2 (0.01)e |  | 0.26 (0.25–0.27) |
| CON × 101–200 DIM | Referentb |  | 0.61 (0.60–0.63) |
| TRT × > 200 DIM | Referentd |  | 0.33 (0.32–0.34) |
| CON × > 200 DIM | Referenta |  | – |

a–fGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S9.** Multivariable generalized linear mixed model showing the effect of treatment, period within the treatment, non-lactating quarter, lactation number, stage of lactation, milking session, log10-transformed somatic cell count (LogSCC), interaction of treatment and lactation number, and interaction of treatment and stage of lactation on forced retract

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | −0.7 (0.03) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.2 (0.02)c |  | 0.49 (0.47−0.52) |
| Period 4 (TRT) | Referentd |  | 0.39 (0.37−0.41) |
| Period 1 (CON) | 0.2 (0.1)a |  | 1.28 (1.23−1.32) |
| Period 3 (CON) | Referentb |  | − |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | −0.1 (0.02) |  | 0.89 (0.87–0.92) |
| Present | Referent |  | – |
| Lactation number |  | <0.0001 |  |
| 1st | −1.3 (0.02)c |  | – |
| 2nd | −0.5 (0.02)b |  | – |
| ≥ 3rd | Referenta |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 1.4 (0.02)a |  | – |
| 101–200 DIM | 0.7 (0.02)b |  | – |
| > 200 DIM | Referentc |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.1 (0.01)b |  | 1.10 (1.07–1.13) |
| Session 2 | 0.2 (0.01)a |  | 1.28 (1.25–1.31) |
| Session 3 | Referentc |  | – |
| LogSCC | −0.1 (0.01) | <0.0001 | 0.89 (0.88−0.91) |
| Treatment × lactation number |  | <0.0001 |  |
| TRT × 1st | −0.1 (0.03)f |  | 0.10 (0.09–0.11) |
| CON × 1st | Referentd |  | 0.27 (0.26–0.28) |
| TRT × 2nd | −0.1 (0.03)e |  | 0.23 (0.22–0.24) |
| CON × 2nd | Referentb |  | 0.62 (0.59–0.64) |
| TRT × 3rd | Referentc |  | 0.42 (0.40–0.44) |
| CON × 3rd | Referenta |  | – |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.2 (0.03)c |  | 1.65 (1.55–1.76) |
| CON × ≤ 100 DIM | Referenta |  | 4.24 (4.02–4.47) |
| TRT × 101–200 DIM | −0.4 (0.04)e |  | 0.67 (0.62–0.72) |
| CON × 101–200 DIM | Referentb |  | 2.12 (2.00–2.24) |
| TRT × > 200 DIM | Referentf |  | 0.47 (0.44–0.52) |
| CON × > 200 DIM | Referentd |  | – |

a–fGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S10.** Multivariable generalized linear mixed model showing the effect of treatment, period within the treatment, non-lactating quarter, lactation number, stage of lactation, milking session, log10-transformed somatic cell count (LogSCC), interaction of treatment and lactation number, and interaction of treatment and stage of lactation on milking liner slip

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.1 (0.03) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | 0.001 |  |
| Period 2 (TRT) | 0.03 (0.02)a |  | 1.07 (1.03−1.12) |
| Period 4 (TRT) | Referentab |  | 1.04 (0.99−1.09) |
| Period 1 (CON) | −0.05 (0.02)c |  | 0.95 (0.91−0.99) |
| Period 3 (CON) | Referentb |  | − |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | −0.9 (0.01) |  | 0.42 (0.41–0.43) |
| Present | Referent |  | – |
| Lactation number |  | <0.0001 |  |
| 1st | −0.6 (0.02)c |  | – |
| 2nd | −0.1 (0.02)b |  | – |
| ≥ 3rd | Referenta |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 0.4 (0.02)a |  | – |
| 101–200 DIM | 0.1 (0.02)b |  | – |
| > 200 DIM | Referentc |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.1 (0.01)b |  | 1.09 (1.06–1.13) |
| Session 2 | 0.1 (0.01)a |  | 1.15 (1.11–1.18) |
| Session 3 | Referentc |  | – |
| LogSCC | 0.2 (0.01) | <0.0001 | 1.23 (1.21−1.25) |
| Treatment × lactation number |  | 0.002 |  |
| TRT × 1st | 0.1 (0.03)d |  | 0.61 (0.57–0.64) |
| CON × 1st | Referente |  | 0.52 (0.49–0.56) |
| TRT × 2nd | 0.03 (0.03)b |  | 0.97 (0.91–1.02) |
| CON × 2nd | Referentc |  | 0.91 (0.86–0.96) |
| TRT × 3rd | Referenta |  | 1.04 (0.99–1.09) |
| CON × 3rd | Referentab |  | – |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.1 (0.03)a |  | 1.59 (1.50–1.69) |
| CON × ≤ 100 DIM | Referentb |  | 1.51 (1.42–1.60) |
| TRT × 101–200 DIM | −0.1 (0.03)c |  | 1.16 (1.09–1.23) |
| CON × 101–200 DIM | Referentc |  | 1.13 (1.06–1.20) |
| TRT × > 200 DIM | Referentc |  | 1.18 (1.10–1.25) |
| CON × > 200 DIM | Referentd |  | – |

a–eGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S11.** Multivariable generalized linear mixed model showing the effect of treatment, non-lactating quarter, lactation number, stage of lactation, milking session, log10-transformed somatic cell count (LogSCC), and interaction of treatment and stage of lactation on milking unit kick-off

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.4 (0.1) |  | – |
| CON | Referent |  | – |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | −1.2 (0.03) |  | 0.30 (0.28–0.32) |
| Present | Referent |  | – |
| Lactation number |  | <0.0001 |  |
| 1st | 1.2 (0.04)a |  | 3.31 (3.07–3.57) |
| 2nd | 0.3 (0.05)b |  | 1.36 (1.24–1.49) |
| ≥ 3rd | Referentc |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 1.5 (0.1)a |  | – |
| 101–200 DIM | 0.6 (0.1)b |  | – |
| > 200 DIM | Referentc |  | – |
| Milking session |  | 0.001 |  |
| Session 1 | 0.05 (0.04)ab |  | 1.06 (0.98–1.14) |
| Session 2 | 0.1 (0.04)a |  | 1.15 (1.07–1.23) |
| Session 3 | Referentb |  | – |
| LogSCC | 0.1 (0.02) | <0.0001 | 1.12 (1.07−1.17) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.3 (0.1)a |  | 4.87 (4.08–5.80) |
| CON × ≤ 100 DIM | Referenta |  | 4.53 (3.80–5.40) |
| TRT × 101–200 DIM | −0.4 (0.1)b |  | 1.89 (1.56–2.29) |
| CON × 101–200 DIM | Referentb |  | 1.83 (1.51–2.22) |
| TRT × > 200 DIM | Referentc |  | 1.51 (1.23–1.86) |
| CON × > 200 DIM | Referentd |  | – |

a–dGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).

**Supplemental Table S12.** Multivariable generalized linear mixed model showing the effect of treatment, period within the treatment, non-lactating quarter, lactation number, stage of lactation, milking session, log10-transformed somatic cell count (LogSCC), and interaction of treatment and stage of lactation on milking unit reattachment

|  |  |  |  |
| --- | --- | --- | --- |
| Variable1 | ß2 (SE) | *P*-value | aOR3(95% CI) |
| Treatment3 |  | <0.0001 |  |
| TRT | 0.5 (0.1) |  | – |
| CON | Referent |  | – |
| Period (Treatment) |  | <0.0001 |  |
| Period 2 (TRT) | 0.4 (0.04)a |  | 2.34 (2.07−2.65) |
| Period 4 (TRT) | Referentb |  | 1.50 (1.30−1.72) |
| Period 1 (CON) | 0.3 (0.05)b |  | 1.39 (1.22−1.59) |
| Period 3 (CON) | Referentc |  | − |
| Non-lactating quarter |  | <0.0001 |  |
| Absent | −0.6 (0.04) |  | 0.58 (0.53–0.62) |
| Present | Referent |  | – |
| Lactation number |  | <0.0001 |  |
| 1st | 0.2 (0.04)a |  | 1.17 (1.09–1.26) |
| 2nd | −0.1 (0.04)c |  | 0.88 (0.81–0.95) |
| ≥ 3rd | Referentb |  | – |
| Stage of lactation |  | <0.0001 |  |
| ≤ 100 DIM | 0.4 (0.06)a |  | – |
| 101–200 DIM | −0.1 (0.06)c |  | – |
| > 200 DIM | Referentb |  | – |
| Milking session |  | <0.0001 |  |
| Session 1 | 0.1 (0.04)a |  | 1.15 (1.07–1.24) |
| Session 2 | −0.1 (0.04)c |  | 0.90 (0.83–0.97) |
| Session 3 | Referentb |  | – |
| LogSCC | 0.2 (0.03) | <0.0001 | 1.18 (1.12−1.24) |
| Treatment × DIM |  | <0.0001 |  |
| TRT × ≤ 100 DIM | −0.3 (0.1)a |  | 2.01 (1.70–2.40) |
| CON × ≤ 100 DIM | Referentb |  | 1.53 (1.28–1.82) |
| TRT × 101–200 DIM | −0.1 (0.1)b |  | 1.53 (1.29–1.82) |
| CON × 101–200 DIM | Referentc |  | 0.92 (0.76–1.11) |
| TRT × > 200 DIM | Referenta |  | 1.83 (1.54–2.16) |
| CON × > 200 DIM | Referentc |  | – |

a–cGroups with different superscript letters differ at a level of *P* < 0.05 in Tukey-Kramer post hoc tests.

1Intercept omitted for clarity.

2Linear regression coefficient.

3Adjusted odds ratio.

4Treatment consisted of 2 different premilking stimulation regimens: flow-responsive vacuum and pulsation with early attachment of milking unit (TRT), and control (CON).