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(54) Title: EARLY LACTATION ADMINISTRATION OF NON-STEROIDAL ANTI-INFLAMMATORY DRUGS TO INCREASE WHOLE-LACTATION MILK YIELD

(57) Abstract: Methods and formulations are provided which can be effectively used to improve milk production and decrease the risk of herd culling in postpartum dairy cattle. The inventive formulations comprise a non-steroidal anti-inflammatory drug, such as meloxicam or sodium salicylate. The inventive method comprises administration of the non-steroidal antiinflammatory drug shortly before, during, and/or after parturition. The inventive methods advantageously provide increased whole-lactation milk yield and increased herd retention with only a very short treatment window.



EARLY LACTATION ADMINISTRATION OF NON-STEROIDAL
ANTI-INFLAMMATORY DRUGS TO INCREASE WHOLE-LACTATION
MILK YIELD

5 CROSS-REFERENCE TO RELATED APPLICATIONS

The present application claims the priority benefit of U.S. Provisional Patent Application Serial No. 62/047,349, filed September 8, 2014, entitled EARLY LACTATION ADMINISTRATION OF NON-STEROIDAL ANTI-INFLAMMATORY DRUGS TO INCREASE WHOLE-LACTATION MILK YIELD AND DECREASE CULLING IN DAIRY
10 CATTLE, incorporated by reference in its entirety herein.

BACKGROUND OF THE INVENTION

Field of the Invention

The present invention is broadly concerned with the use of non-steroidal anti-inflammatory drugs on postpartum dairy cows to increase milk production and decrease herd
15 culling.

Description of Related Art

There is growing evidence that inflammation is common in early lactation dairy cows. Although some degree of inflammation is probably necessary for immune system activation, excess inflammation has been linked to a number of negative outcomes. In particular,
20 inflammation in early lactation has been repeatedly linked to decreased whole-lactation milk production. One diagnostic marker of inflammation is a decline in blood paraoxanase, and transition cows with low paraoxanase concentrations produced 24% less milk (4,300 lbs less) over 305 days than those in the highest quartile for paraoxanase (Bionaz et al., 2007). In another
25 study, blood plasma haptoglobin concentrations (induced during inflammation) greater than 1.1 g/L were associated with a 2,100 lb decrease in 305-day mature equivalent milk yield as well as decreased risk of conception (Huzzey et al., 2012). The use of anti-inflammatory treatment in early lactation to increase milk production has been previously described (Bertoni et al., 2004; Trevisi and Bertoni, 2008). However, milk produced by cows while being treated with anti-
30 inflammatory drugs must be disposed of and these dairy cattle are often culled. What is needed is a short-term treatment that increases milk yield and reduces culling.

SUMMARY OF THE INVENTION

In one embodiment of the present invention, there is provided methods of increasing milk production of a postpartum dairy cow. The method comprises administering to the cow an effective amount of a non-steroidal anti-inflammatory drug less than about 1 month after parturition and milking the cow. The non-steroidal anti-inflammatory drug is selected from the group consisting of propionic acid derivatives, acetic acid derivatives, enolic acid derivatives, nicotinic acid derivatives, tolafenamic acid derivatives, anthranilic acid derivatives, selective COX-2 inhibitors, sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof.

In another embodiment, there is provided methods of increasing milk production of a postpartum dairy cow. The method comprises administering to the cow an effective amount of a non-steroidal anti-inflammatory drug and milking the cow. The drug is first administered to the cow less than about 1 month after parturition and the administering is discontinued less than 1 week after the first administration.

In yet another embodiment, there is provided methods of preventing a postpartum cow from being culled from the herd. The method comprises administering to the cow an effective amount of a non-steroidal anti-inflammatory drug less than about 1 month after parturition.

In still another embodiment, there is provided a use of a non-steroidal anti-inflammatory drug to improve milk production in second or greater parity dairy cows.

Additional advantages of the invention will be appreciated based upon the figures and the detailed description of the preferred embodiments below.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows whole-lactation milk responses following early-lactation treatment with placebo (CON), meloxicam (MEL), or sodium salicylate (SAL);

Figure 2 is a graph showing herd survival analysis following early-lactation treatment with placebo (CON), meloxicam (MEL), or sodium salicylate (SAL); and

Figure 3 shows plasma constituent responses following early-lactation treatment with placebo (CON), meloxicam (MEL), or sodium salicylate (SAL).

DETAILED DESCRIPTION OF THE INVENTION

In more detail, the present invention is concerned with methods of improving milk yield in dairy cattle and/or decreasing dairy cattle herd culling. More specifically, the invention relates to the use of non-steroidal anti-inflammatory drugs to increase whole-lactation milk production and/or increasing herd retention of postpartum dairy cows. Advantageously, the present invention is effective in providing long-term results with only short-term treatment to dairy cows shortly before, during, and/or after parturition. For example, in certain embodiments, the present invention can provide increased milk production and/or decreased culling rates over the span of at least about 305 days with treatments comprising an effective amount of a non-steroidal anti-inflammatory drug beginning less than about 1 month after parturition and lasting less than 1 week. As used herein, an "effective amount" refers to an amount capable of providing bioavailable levels of the active compound (i.e., non-steroidal anti-inflammatory drug) sufficient to achieve the desired performance improvement. "Parturition" or "calving" is a three stage process of calf birthing comprising dilation of the cervix, delivery of the calf, and delivery of the placenta. As used herein, "after parturition" refers to the period of time beginning after the third parturition stage (delivery of the placenta) is completed. In preferred embodiments, methods of the present invention are advantageously adapted for use on commercial dairy farms.

In the invention, an effective amount of a non-steroidal anti-inflammatory drug is administered to dairy cows within a relatively short time span before, during, and/or after parturition. In certain embodiments, the non-steroidal anti-inflammatory drug is first administered to a dairy cow less than about 1 month after parturition, preferably less than about 1 week after parturition, more preferably less than about 3 days after parturition, and most preferably less than about 36 hours after parturition. In certain other embodiment, the non-steroidal anti-inflammatory drug may be first administered before the parturition process begins (i.e., before dilation of the cervix), for example, up to about 1 week before parturition, preferably up to about 3 days before parturition, or more preferably up to about 1 day before parturition. In other embodiments, the non-steroidal drug may be first administered during parturition, for example, after dilation of the cervix or delivery of the calf but before delivery of the placenta. The non-steroidal anti-inflammatory drug treatment may be administered as a single dose or as multiple doses spread out over multiple days. For example, in some embodiments, the non-

steroidal anti-inflammatory drug may be administered as a single dose less than about 1 month, less than about 1 week, less than about 3 days, or less than about 36 hours after parturition (or up to about 1 week, 3 days, or 1 day before parturition, or during parturition). In some such embodiments, for example, a single dose may be administered less than about 1 week after parturition and no subsequent dose is administered less than 1 week after parturition. In other
5 embodiments, the non-steroidal anti-inflammatory drug may be administered as a first dose less than about 1 month, less than about 1 week, less than about 3 days, or less than about 36 hours after parturition (or up to about 1 week, 3 days, or 1 day before parturition, or during parturition) and then additional doses daily for up to 1 week, up to 6 consecutive days, up to 5 consecutive
10 days, or up to 3 consecutive days after the first administration. In certain preferred embodiments, the non-steroidal anti-inflammatory drug is not administered for more than 3 consecutive days after the first administration, although in some embodiments, administration may continue beyond 3 days. For example, in some embodiments, administration of the non-steroidal anti-inflammatory drug can occur up to a week after parturition or even up to a month
15 after parturition. The non-steroidal anti-inflammatory drug is preferably in a form suitable for oral or parenteral administration, including without limitation, tablets, granules, capsules, liquids, top-dress preparations, suspensions, bolus, drenches, solutions, topical pour-ons, pre-mixes, or sustained-release implants. Rectal suppositories may also be used.

Non-steroidal anti-inflammatory drugs for use in accordance with the present invention
20 are a class of non-narcotic analgesics known to reduce the production of prostaglandins in a body's cells by inhibiting cyclooxygenase (COX) enzymes. Prostaglandins are produced by COX enzymes to promote inflammation in response to physical stress placed on body tissues. In dairy cows, for example, COX enzymes can produce prostaglandins causing systemic metabolic inflammation following parturition. Non-steroidal anti-inflammatory drugs for use in
25 accordance with the present invention include salicylates, propionic acid derivatives (e.g., ibuprofen), acetic acid derivatives, enolic acid (oxicam) derivatives, nicotinic acid derivatives (flunixin), anthranilic acid derivatives (fenamates, tolfenamic acid and derivatives), selective COX-2 inhibitors (coxibs), sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof. In certain embodiments, salicylates (such as sodium salicylate) are not used.
30 Therefore, in such embodiments, the non-steroidal anti-inflammatory drug is selected from the group consisting of propionic acid derivatives, acetic acid derivatives, enolic acid derivatives,

nicotinic acid derivatives, tolfenamic acid derivatives, anthranilic acid derivatives, selective COX-2 inhibitors, sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof. In certain other embodiments, however, salicylates may be used. In those such
5 salicylic acid and salts and derivatives thereof, propionic acid derivatives, acetic acid derivatives, enolic acid derivatives, nicotinic acid derivatives, tolfenamic acid derivatives, anthranilic acid derivatives, selective COX-2 inhibitors, sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof.

In a preferred embodiment, the non-steroidal anti-inflammatory drug is enolic acid
10 derivative such as meloxicam or a pharmaceutically acceptable salt thereof. A pharmaceutically acceptable "salt" of meloxicam includes acid addition salts, such as the meglumine, sodium, potassium, tartrate, or ammonium salts. Meloxicam and suitable salts are disclosed in U.S. Patent No. 4,233,299, incorporated by reference herein to the extent not inconsistent with the present invention. The term "pharmaceutically acceptable," as used herein, is meant to include
15 compounds that are not biologically or otherwise undesirable, i.e., the compound may be administered without causing any undesirable biological effects or interacting in a deleterious manner or interfering with any of the other components of the composition in which it is contained. The meloxicam or pharmaceutically acceptable salt thereof is administered to the cow in an amount sufficient to provide meloxicam levels (independent of salt, if any) of from
20 about 0.5 mg to about 10 mg of meloxicam per kg of body weight of the cow, preferably from about 0.6 mg/kg to about 5 mg/kg of body weight of the cow, more preferably from about 0.7 mg/kg to about 3 mg/kg of body weight of the cow, even more preferably from about 0.8 mg/kg to about 2 mg/kg of body weight of the cow, and most preferably about 1 mg/kg of body weight of the cow. Thus, it will be appreciated that in the case of meloxicam salts, for example, the
25 formulation may be administered in amounts greater than the above ranges to provide sufficient levels of the active meloxicam (e.g., 4.1 mg/kg meloxicam tartrate is equivalent to 2.8 mg/kg meloxicam base). In certain embodiments, meloxicam or a pharmaceutically acceptable salt thereof is administered to the cow in a single dose less than about 1 month after parturition, preferably less than about 1 week after parturition, more preferably less than about 3 days after
30 parturition, and even more preferably less than about 36 hours after parturition. In certain other embodiments, the single dose may be administered before parturition (up to about 1 week, up to

3 days, or up to 1 day before parturition) or during parturition. The single dose is administered to said cow in an effective amount, for example, providing an amount of active meloxicam of from about 300 mg to about 6,000 mg, preferably from about 400 mg to about 3,000 mg, more preferably from about 500 mg to about 1,500 mg, even more preferably from about 600 mg to about 1,000 mg, and most preferably about 675 mg.

In another preferred embodiment, the non-steroidal anti-inflammatory drug is salicylic acid or a pharmaceutically acceptable derivative or salt thereof. For example, in certain embodiments the non-steroidal anti-inflammatory drug is selected from the group consisting of salicylic acid, sodium salicylate, methyl salicylate, salsalate, diflunisal, or acetylsalicylic acid.

The salicylic acid or a pharmaceutically acceptable derivative or salt thereof is administered to the cow in an amount sufficient to provide salicylate levels of from about 25 mg to about 500 mg of salicylate per kg of body weight of the cow, preferably from about 100 mg/kg to about 300 mg/kg of body weight of the cow, more preferably from about 150 mg/kg to about 200 mg/kg of body weight of the cow, and most preferably about 160 mg/kg of body weight of the cow. The salicylic acid or a pharmaceutically acceptable derivative or salt thereof may be administered to the cow as a single dose less than about 1 month, 1 week, 3 days, or 36 hours after parturition or as multiple daily doses for up to 1 week, up to 6 consecutive days, up to 5 consecutive days, or up to 3 consecutive days with treatment beginning less than about 1 month, 1 week, 3 days, or 36 hours after parturition. In certain preferred embodiments, for example, sodium salicylate or other salicylate salt is administered to said cow in a daily dosage of from about 25 g to about 400 g of dissolved sodium salicylate, preferably from about 50 g to about 250 g of dissolved sodium salicylate, more preferably from about 100 g to about 175 g of dissolved sodium salicylate, and more preferably about 125 g of dissolved sodium salicylate, for 3 or more consecutive days. In some embodiments, the first daily dose is administered less than about 1 month, 1 week, 3 days, or 36 hours after parturition and/or administration is discontinued less than 1 week, less than 6 days, less than 5 days, or less than 4 days after the first administration. In certain other embodiments, the single dose or first daily dose may be administered before parturition (up to about 1 week, up to about 3 days, or up to about 1 day before parturition) or during parturition.

The non-steroidal anti-inflammatory drug can be administered directly or as part of a veterinary formulation comprising the non-steroidal anti-inflammatory drug and a carrier. The term "carrier," as used herein, means one or more compatible base compositions with which the

active ingredient (e.g., meloxicam, salicylate, etc.) is combined to facilitate the administration of ingredient, and which is suitable for administration to a dairy cow. Such preparations may also routinely contain salts, buffering agents, preservatives, and optionally other therapeutic ingredients. The carrier would naturally be selected to minimize any degradation of the active ingredient and to minimize any adverse side effects in the subject, as would be well known to one of ordinary skill in the art. For example, the veterinary formulation can be a liquid system for oral or parenteral administration comprising the non-steroidal anti-inflammatory drug dispersed or dissolved in a carrier. Exemplary carriers for this embodiment, depending upon the method of administration, would include water, oil, water-in-oil or oil-in-water emulsion, milk, milk replacer, propylene glycol, polyethylene glycol, n-methyl-2-pyrrolidone, feed top-dress, pre-mix, or bolus. The veterinary formulation can also be a solid system for oral administration comprising non-steroidal anti-inflammatory drug dispersed in a carrier. Exemplary carriers in this embodiment can include binders (e.g. gelatin, cellulose, talc, hydroxyethylcellulose, carboxyethylcellulose, silicon dioxide, hypromellose, magnesium stearate, polydextrose, titanium dioxide), sweeteners (e.g., glucose, sucrose), and/or flavoring agents. It will also be appreciated that a solid veterinary formulation containing non-steroidal anti-inflammatory drug can be first dissolved in a liquid medium to create a solution or suspension, which is then administered to the cow.

In certain methods in accordance with the present invention, the non-steroidal anti-inflammatory drug is administered to a dairy cow orally or parenterally. That is, in such embodiments, the non-steroidal anti-inflammatory drug is administered systemically as opposed to being injected directly into a site of inflammation on the cow. Parenteral administration in the present invention includes subcutaneous, intramuscular, transdermal, or intravenous injection/administration. Oral administration can include separate administration of the formulation (e.g., hand feeding of tablets, etc., or administration of suspension via gavage tube) or inclusion of the non-steroidal anti-inflammatory drug in the feed, water, or milk/milk replacer to be fed to the cow. In certain preferred embodiments, the non-steroidal anti-inflammatory drug advantageously does not need to be used in a daily dosing regimen to be effective. Rather, the non-steroidal anti-inflammatory drug is administered as a single dose. More specifically, in such preferred embodiments, the non-steroidal anti-inflammatory drug is generally administered to the cow as a single dose shortly before, during, or after (less than about 1 month, 1 week, 3

days, or 36 hours) parturition. For example, where the cow has been administered a non-steroidal anti-inflammatory drug, the initial dose is preferably not followed up with a subsequent (or daily) administration of the drug. In certain other preferred embodiments, however, the non-steroidal anti-inflammatory drug is administered as multiple daily doses beginning shortly
5 before, during, or after (less than about 1 month, preferably less than about 1 week, more preferably less than about 3 days, or even more preferably less than about 36 hours) parturition. In such embodiments, the non-steroidal anti-inflammatory drug may be first administered before parturition (up to about 1 week, up to about 3 days, or up to about 1 day before parturition), during parturition, or after parturition (less than about 1 month, 1 week, 3 days, or 36 hours after
10 parturition) and continue daily for up to about 1 week, preferably up to 6 consecutive days, more preferably up to 5 consecutive days, and most preferably up to 3 consecutive days. In such embodiments, the daily administration is discontinued after 1 week, 6 consecutive days, 5 consecutive days, or 3 consecutive days of administration. In certain embodiments, the non-steroidal anti-inflammatory drug is administered to the cow daily as an oral drench comprising
15 an aqueous solution of the non-steroidal anti-inflammatory drug.

According to a preferred aspect, the present invention is effective for increasing milk production in postpartum dairy cows. In one method, an effective amount of a non-steroidal anti-inflammatory drug is administered to a dairy cow shortly before, during, or after parturition, as described above. The dairy cow is then milked, for example, by hand milking, vacuum bucket
20 milking, milking pipeline, milking parlor (i.e., herringbone, parallel, or rotary parlors), robotic milking, or any other milking method generally known in the art. Advantageously, dairy cows treated in accordance with the methods of the present invention show whole-lactation (305-day mature equivalent) milk production that is increased by at least about 6%, more preferably by at least about 7%, or more preferably by at least about 9%, when compared to untreated dairy cows.
25 Additionally, cows treated in accordance with the inventive methods show weekly milk yield (305-day mature equivalent) production that is increased by at least about 8%, preferably at least 10%, or more preferably at least about 12% as compared to untreated cows. In some embodiments, the increased milk yield is seen after about one month after the first administration of the non-steroidal anti-inflammatory drug and remaining for at least a year or at least another
30 44 weeks of lactation. This aspect of the invention is effective on dairy cows of various ages and parity, as well as on both healthy cows and cows suffering from postpartum mastitis or other

illness. For example, the inventive methods are advantageously effective in increasing milk yield in second (or greater) parity dairy cows. In certain additional embodiments, the inventive methods are effective in increasing milk yield in first (or greater) parity dairy cows.

In another preferred aspect, the present invention is effective for increasing retention (i.e.,
5 decreasing culling) in a dairy cow herd. Dairy cows treated with methods in accordance with the present invention experience at least about 10%, preferably at least about 15%, or more preferably at least about 20% greater herd survival rate compared to untreated cows. Advantageously, the methods of the present invention decrease herd culling due to diseases such as respiratory disease, displaced abomasum, and metabolic disorders. Therefore, this aspect of
10 the present invention is directed to preventing a postpartum cow from being culled from the herd.

In all aspects, the methods of the present invention advantageously have no substantial negative effect on the body condition over the lactation period, as determined by body condition scoring. Body condition scoring is a technique known in the art to provide an objective indication of the amount of fat cover on a dairy cow. A cow is assigned a score based on the
15 amount of fat observed on various skeletal parts of the cow. The scoring observer assigns a score ranging from 1.0 to 5.0, in increments of 0.1, based on the observed fat on the cow. Additionally, the methods have no substantial negative effect on plasma non-esterified fatty acids concentration or plasma paraoxonase activity of said cow during the lactation period.

20

EXAMPLE

The following example sets forth methods in accordance with the invention. It is to be understood, however, that this example is provided by way of illustration and nothing therein should be taken as a limitation upon the overall scope of the invention.

In this Example, there is provided a study of the effects of a treatment protocol on
25 commercial dairy farm cattle. Specifically, the study analyzed the effects of non-steroidal anti-inflammatory drug treatments on milk production and herd retention. Two treatments were studied, one using sodium salicylate as the active ingredient and the other using meloxicam as the active ingredient.

METHODS AND MATERIALS

30 *Animals & Treatments*

Cows from a commercial dairy (n = 153) were enrolled in the study 12-36 hours after calving. Cows assigned to sodium salicylate (SS) treatment received 1 placebo bolus containing casein on day 1 of treatment and an oral drench containing 125 g/day of SS in 375 mL of water for 3 consecutive days beginning on day 1 of treatment. Cows assigned to meloxicam (M) treatment received 675 mg of meloxicam as a bolus on day 1 of treatment as well as 3 consecutive daily drenches of 375 mL of water. Control cows (CON) received a placebo bolus on day 1 and water drenches for 3 days. Cows were blocked by mastitis at parturition (CON = 1, M = 2, SS = 2), breed (CON = 6, M = 6, SS = 4 crossbred; all others were Holstein), dystocia (calving difficulty score ≥ 3 ; CON = 5, M = 5, SS = 6), and twin births (CON = 4, M = 4, SS = 3) and were sequentially assigned to treatment within block. Milk from treated cows was discarded for the first 7 days in milk, and M cows were not allowed to be slaughtered until 21 days in milk. Cows on the M treatment that were not slaughtered, but otherwise would have been, were treated as culled in study analysis and data after that point in time were not used for analysis.

Farm employees, who made all management and culling decisions, were blinded to treatment. Cows on the study were milked 4x/day for the first 28 days of lactation, and 3x/day for the remainder of the lactation.

Data Collection & Analysis

Milk yield data were recorded electronically at each milking and averaged by week over 44 weeks of lactation. Additionally, monthly Dairy Herd Improvement Association (DHIA) testing and determination of 305-day yields of milk, fat, and protein were utilized in an independent analysis. Weekly milk data were analyzed with fixed effects of block, parity, treatment, week, and treatment by week interaction along with the random effect of cow. The model accounted for repeated measures over time with autoregressive covariance structures. DHIA data on 305-day yields were analyzed with models including blocking factors as well as animal-level genetic predictions for these traits as a covariate factor. Blocking factors and interactions were tested and removed from models when $P > 0.10$, and treatment contrasts were evaluated using the Student's t test with significance declared at $P < 0.05$.

Removal rate from the herd was evaluated by Cox regression proportional hazard analysis, and incidences of diseases were tested by Fisher's exact test. Treatment, breed, dystocia, twin births, and mastitis at enrollment were accounted for in the model.

Body condition was evaluated on day 3 of treatment and approximately 2, 5, and 8 mo post-treatment. Body condition score was recorded as the average of responses from at least 3 independent observers.

Blood samples were collected via the coccygeal vein on the first and last day of treatment and 7 days after the completion of treatment. Plasma was collected and stored at -20°C until analyzed for glucose by a colorimetric kit (kit #439-90901; Wako Chemicals USA Inc.), non-esterfied fatty acids (NEFA) using an enzymatic colorimetric procedure (NEFA-HR; Wako Chemicals USA Inc., Richmond, VA), and β -hydroxybutyrate (BHBA) using an enzymatic reaction (kit #H7587-58; Pointe Scientific Inc., Canton, MI). Haptoglobin was measured by the method of Cooke and Arthington (2012), a colorimetric technique that uses differences in peroxidase activity to measure haptoglobin-haemoglobin complexing. Absorbance was measured with a spectrophotometer (PowerWave XS; BioTek Instruments Inc., Winooski, VT) and calculations were performed using Gen5 software (BioTek Instruments Inc.). Paraoxonase was measured by the method of Ferré et al. (2002).

RESULTS

Whole-lactation (305-day mature equivalent) yields of milk and milk protein were significantly increased by both meloxicam and sodium salicylate treatments ($P < 0.05$), with 6 - 9% increases observed. Milk fat yield was not significantly different across treatments. Weekly milk yield analysis resulted in a similar 10 - 12% increase in milk yield after meloxicam and sodium salicylate treatment (both $P < 0.05$), with an effect first appearing in week 4 of lactation and remaining throughout the remainder of the 44-week lactation. Results of the milk yield study are shown in graphs A-B of Figure 1. In the graphs, values are means $\pm$ standard error (SEM). Graph A shows daily milk yield data compiled by week and analyzed with repeated measures (pooled SEM = 2.4 kg/d). Asterisks (*) indicate that non-steroidal anti-inflammatory drug treatments showed significant statistical difference from the control (CON) ($P < 0.05$, $n = 51$). Graph B shows 305-day mature equivalent component yields, analyzed in a model that accounts for genetic effects. Asterisks (*) indicate that non-steroidal anti-inflammatory drug treatments showed significant statistical difference from the control (CON) ($P < 0.05$, $n = 39-42$).

A total of 47 cows on the study left the herd during the first 300 days after parturition, and cows treated with meloxicam were ($P = 0.04$) removed from the herd at a slower rate than

control cows (76% survival vs. 61% survival). Treatment did not affect the risk of leaving the herd due to mastitis, low milk production, injury, lameness, or death (all $P > 0.1$). However, meloxicam treatment tended to decrease the risk of culling due to other diseases—including respiratory disease, displaced abomasum, and suspected metabolic disorders—compared to the control ($n = 7$ CON vs. 2 M; $P = 0.09$). These results indicate that in addition to elevated milk production, postpartum administration of meloxicam may have beneficial effects on dairy cow longevity. Results of the herd retention study are shown in the graph of Figure 2. The graph shows retention in the herd by treatment: 30, 38, and 35 cows remained in the herd at 365 days post-treatment for CON, MEL, and SAL, respectively. In particular, meloxicam (MEL) tended to delay removal from the herd relative to the control (CON) ($P = 0.06$, Wilcoxon chi-square test).

Results of the body condition score analysis showed no meaningful difference between cows treated with meloxicam or sodium salicylate and the control cows. Results of the blood sample analyses are presented in graphs A-D of Figure 3. Unless otherwise noted in the graphs, values represent the overall mean ($\pm$ standard error (SEM), $n = 50$ -51) across samples collected at 3 and 10 days-in-milk, and means with uncommon superscripts differ ($P < 0.05$). Graph A shows plasma glucose concentration increased by meloxicam (MEL) in parity 3+ cows. Graph B shows that treatment interacted with time to alter plasma β -hydroxybutyrate (BHBA) concentration. Asterisks (*) indicate that sodium salicylate (SAL) showed significant statistical difference from meloxicam (MEL) at day 7 post-treatment ($P = 0.02$). Graph C shows the plasma non-esterfied fatty acids (NEFA) concentration was not altered by treatment, but sodium salicylate (SAL) increased haptoglobin concentration compared to the control (CON). Graph D shows plasma paraoxonase activity unaltered by treatment. Notably, treatment appeared to have substantially no negative effect on non-esterfied fatty acids (NEFA) plasma concentration or plasma paraoxonase activity.

Claims:

1. A method of increasing milk production of a postpartum dairy cow,
5 comprising:
administering to said cow an effective amount of a non-steroidal anti-inflammatory drug less than about 1 month after parturition, wherein said non-steroidal anti-inflammatory drug is selected from the group consisting of propionic acid derivatives, acetic acid derivatives, enolic acid
10 derivatives, nicotinic acid derivatives, tolfenamic acid derivatives, anthranilic acid derivatives, selective COX-2 inhibitors, sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof; and milking said cow, wherein said cow has increased milk production.
- 15 2. The method of claim 1, wherein a first dose of said non-steroidal anti-inflammatory drug is first administered to said cow less than about 1 week after parturition.
3. The method of claim 2, wherein said non-steroidal anti-inflammatory drug is administered as a single dose less than about 1 week after parturition, wherein no subsequent
20 dose is administered less than about 1 week after parturition.
4. The method of claim 2, wherein said non-steroidal anti-inflammatory drug is administered as multiple daily doses for up to 6 consecutive days after the first administration.
- 25 5. The method of claim 4, wherein administration of daily doses is discontinued 3 days after the first administration.
6. The method of claim 1, wherein said non-steroidal anti-inflammatory drug is in the form of tablet, capsule, liquid, granule, top-dress, suspension, bolus, drench, solution,
30 topical pour-on, sustained-release implant, or rectal suppository.

7. The method of claim 1, wherein said non-steroidal anti-inflammatory drug is administered orally, parenterally, or both.

8. The method of claim 7, wherein said drug is administered parenterally and
5 wherein said drug is administered subcutaneously, transdermally, intramuscularly, or intravenously.

9. The method of claim 7, wherein said drug is administered orally in the form of a bolus or drench.
10

10. The method of claim 1, wherein said non-steroidal anti-inflammatory drug is meloxicam or a pharmaceutically acceptable salt thereof.

11. The method of claim 10, wherein said meloxicam or pharmaceutically
15 acceptable salt thereof is administered as a single dosage of about 300 to about 6,000 milligrams.

12. The method of claim 10, wherein said meloxicam or pharmaceutically acceptable salt thereof is administered to said cow in an amount sufficient to provide an amount of active meloxicam of about 0.5 to about 10 mg/kg body weight of said cow.
20

13. The method of any of claims 1 to 12, wherein said method has substantially no negative effect on plasma non-esterified fatty acids concentration or plasma paraoxonase activity of said cow.

25 14. A method of increasing milk production of a postpartum dairy cow, comprising:

administering to said cow an effective amount of a non-steroidal anti-inflammatory drug, wherein said drug is first administered to said cow less than about 1 month after parturition and wherein said administering is
30 discontinued less than 1 week after the first administration; and milking said cow, wherein said cow has increased milk production.

15. The method of claim 14, wherein said non-steroidal anti-inflammatory drug is selected from the group consisting of salicylic acid and salts and derivatives thereof, propionic acid derivatives, acetic acid derivatives, enolic acid derivatives, anthranilic acid
5 derivatives, selective COX-2 inhibitors, sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof.

16. The method of claim 14, wherein said non-steroidal anti-inflammatory drug is administered as a single dose less than about 1 week after parturition, wherein no
10 subsequent dose is administered less than about 1 week after parturition.

17. The method of claim 14, wherein said non-steroidal anti-inflammatory drug is administered as multiple daily doses for up to 6 consecutive days after the first administration.

15

18. The method of claim 17, wherein administration of daily doses is discontinued 3 days after the first administration.

19. The method of claim 14, wherein said non-steroidal anti-inflammatory
20 drug is in the form of tablet, capsule, liquid, granule, top-dress, suspension, bolus, drench, solution, topical pour-on, sustained-release implant, or rectal suppository.

20. The method of claim 14, wherein said non-steroidal anti-inflammatory drug is administered orally, parenterally, or both.

25

21. The method of claim 20, wherein said drug is administered parenterally and wherein said drug is administered subcutaneously, transdermally, intramuscularly, or intravenously.

22. The method of claim 20, wherein said drug is administered orally in the form of a bolus or drench.

30

23. The method of claim 14, wherein said non-steroidal anti-inflammatory drug is meloxicam or a pharmaceutically acceptable salt thereof.

5

24. The method of claim 23, wherein said meloxicam or pharmaceutically acceptable derivative or salt thereof is administered as a single dosage of about 300 to about 6,000 milligrams.

10

25. The method of claim 23, wherein said meloxicam or pharmaceutically acceptable derivative or salt thereof is administered to said cow in an amount sufficient to provide an amount of active meloxicam of about 0.5 to about 10 mg/kg body weight of said cow.

15

26. The method of claim 14, wherein said non-steroidal anti-inflammatory drug is salicylic acid or a pharmaceutically acceptable derivative or salt thereof.

27. The method of claim 26, wherein said non-steroidal anti-inflammatory drug is sodium salicylate.

20

28. The method of claim 27, wherein said sodium salicylate is administered as multiple daily doses of about 25 to about 400 grams for at least 3 consecutive days.

29. The method of claim 26, wherein said salicylic acid or a pharmaceutically acceptable derivative or salt thereof is administered to said cow in an amount sufficient to provide an amount of active salicylate of about 50 to about 500 mg/kg body weight of said cow.

25

30. The method of any of claims 21 to 36, wherein said method has substantially no negligible effect on plasma non-esterified fatty acids concentration or plasma paraoxonase activity of said cow.

30

31. A method of preventing a postpartum cow from being culled from the herd, comprising administering to said cow an effective amount of a non-steroidal anti-inflammatory drug less than about 1 month after parturition.

5 32. The method of claim 31, wherein said non-steroidal anti-inflammatory drug is selected from the group consisting of salicylic acid and salts and derivatives thereof, propionic acid derivatives, acetic acid derivatives, enolic acid derivatives, nicotinic acid derivatives, tolafenamic acid derivatives, anthranilic acid derivatives, selective COX-2 inhibitors, sulfonnanilides, clonixin, clonixeril, licofelone, H-harpagide, and mixtures thereof.

10 33. The method of claim 31, wherein a dose of said non-steroidal anti-inflammatory drug is first administered to said cow less than about 1 week after parturition.

15 34. The method of claim 33, wherein said non-steroidal anti-inflammatory drug is administered as a single dose less than about 1 week after parturition, wherein no subsequent dose is administered less than about 1 week after parturition.

20 35. The method of claim 33, wherein said non-steroidal anti-inflammatory drug is administered as multiple daily doses for up to 6 consecutive days after the first administration.

36. The method of claim 35, wherein administration of daily doses is discontinued 3 days after the first administration.

25 37. The method of claim 31, wherein said non-steroidal anti-inflammatory drug is in the form of tablet, capsule, liquid, granule, top-dress, suspension, bolus, drench, solution, topical pour-on, sustained-release implant, or rectal suppository.

30 38. The method of claim 31, wherein said non-steroidal anti-inflammatory drug is administered orally, parenterally, or both.

39. The method of claim 38, wherein said drug is administered parenterally and wherein said drug is administered subcutaneously, transdermally, intramuscularly, or intravenously.

5 40. The method of claim 38, wherein said drug is administered orally in the form of a bolus or drench.

41. The method of claim 31, wherein said non-steroidal anti-inflammatory drug is meloxicam or a pharmaceutically acceptable salt thereof.

10 42. The method of claim 41, wherein said meloxicam or pharmaceutically acceptable derivative or salt thereof is administered as a single dosage of about 300 to about 6,000 milligrams.

15 43. The method of claim 41, wherein said meloxicam or pharmaceutically acceptable derivative or salt thereof is administered to said cow in an amount sufficient to provide an amount of active meloxicam of about 0.5 to about 10 mg/kg body weight of said cow.

20 44. The method of claim 31, wherein said non-steroidal anti-inflammatory drug is salicylic acid or a pharmaceutically acceptable derivative or salt thereof.

25 45. The method of claim 44, wherein said non-steroidal anti-inflammatory drug is sodium salicylate.

46. The method of claim 45, wherein said sodium salicylate is administered as multiple daily doses of about 25 to about 400 grams for at least 3 consecutive days.

47. The method of claim 44, wherein said salicylic acid or a pharmaceutically acceptable derivative or salt thereof is administered to said cow in an amount sufficient to provide an amount of active salicylate of about 50 to about 500 mg/kg body weight of said cow.

5 48. Use of a non-steroidal anti-inflammatory drug to improve milk production in second or greater parity dairy cows.

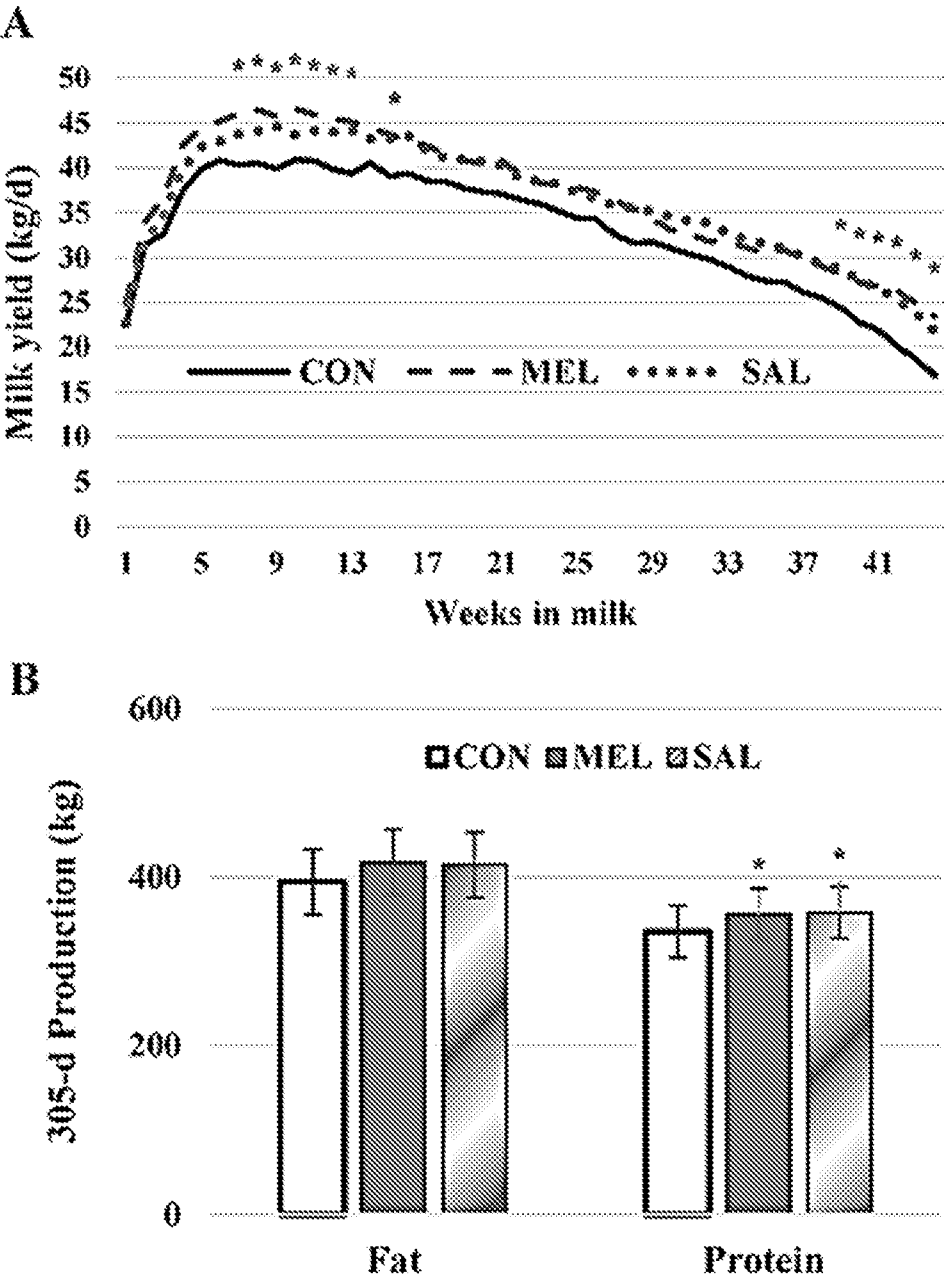


FIGURE 1

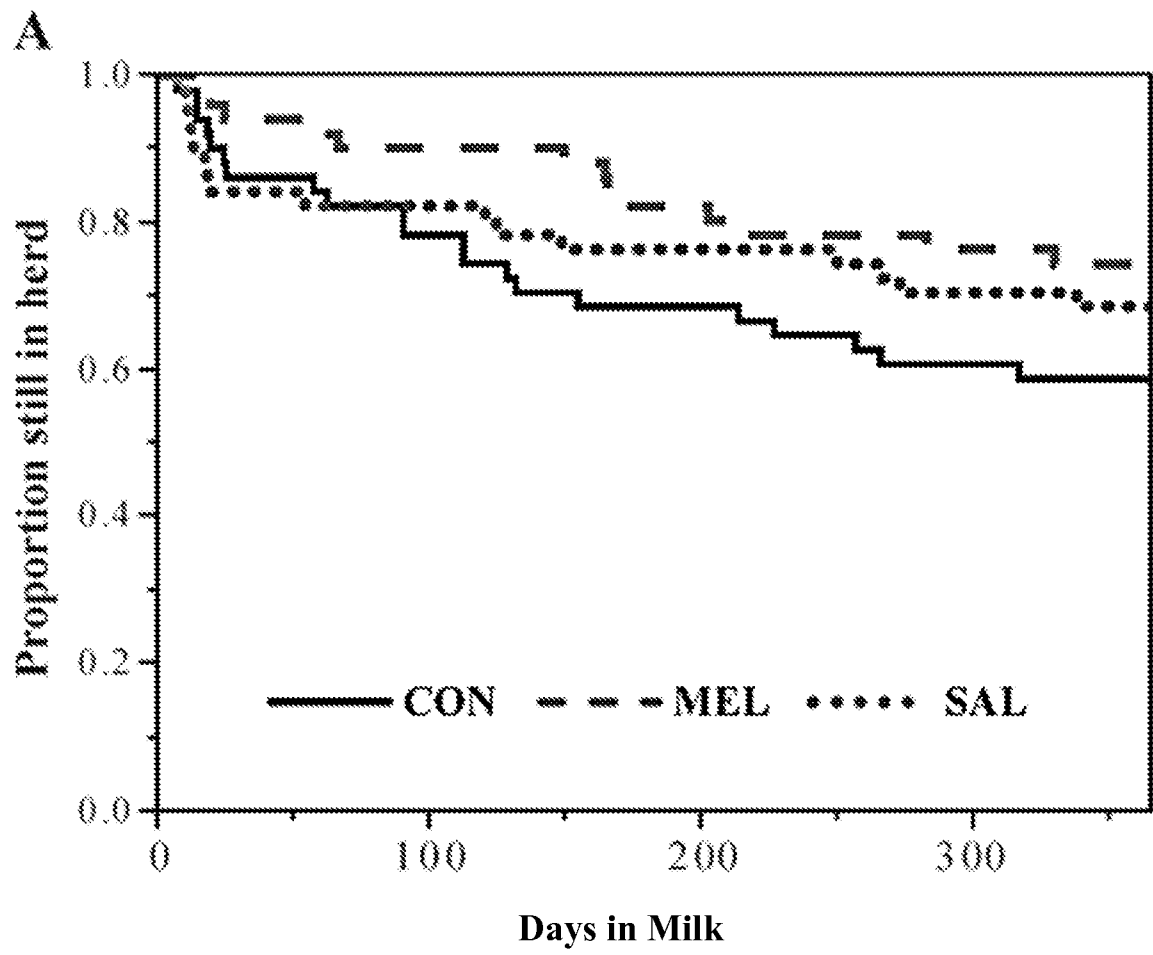


FIGURE 2

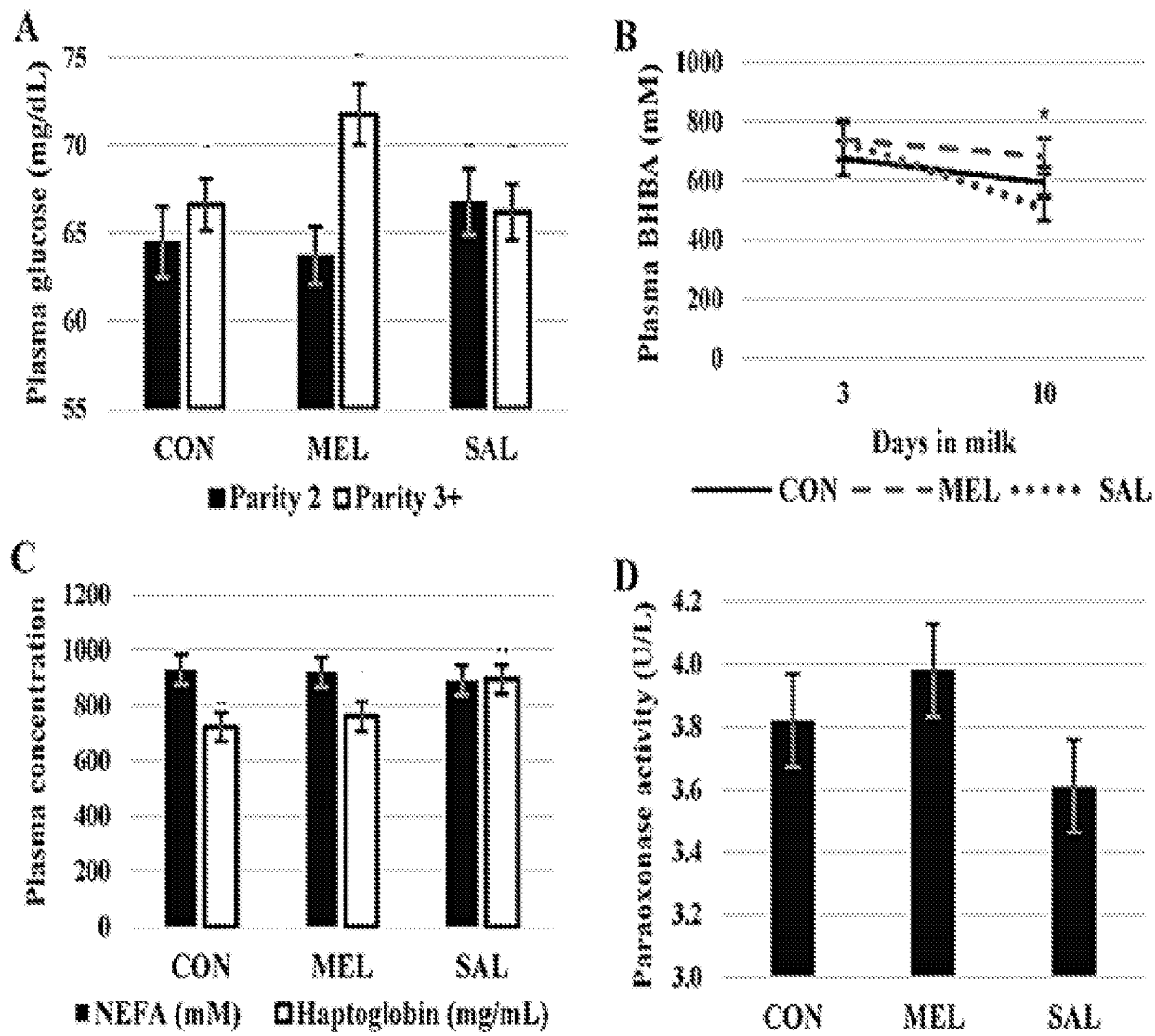


FIGURE 3

INTERNATIONAL SEARCH REPORT

International application No.

PCT/US 15/48977

A. CLASSIFICATION OF SUBJECT MATTER IPC(8) - A61K 38/00 (2015.01) CPC - A23V 2002/00; A23L 1/296; A23L 1/3053 According to International Patent Classification (IPC) or to both national classification and IPC		
B. FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) IPC(8) - A61K 38/00 (2015.01) CPC - A23V 2002/00; A23L 1/296; A23L 1/3053 Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched UC - 514/5.5, 119/174, 514/11.3 Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) Minesoft Patbase, Google Scholar, keywords: increasing milk production postpartum dairy coW administering non-steroidal antiinflammatory drug propionic acid acetic acid derivatives, enolic acid nicotinic acid tolfenamic acid, anthranilic acid derivatives selective COX-2 inhibitors sulfonamides		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X ----	Carpenter et al., 'Effect of Postpartum Treatment with Non-Steroidal Anti-Inflammatory Drugs (NSAID) on Reproductive Performance and Removal from the Herd in Dairy Cattle through Mid-Lactation', conference proceeding, July 21, 2014, URL: https://asas.confex.com/asas/jam2014/webprogram/Paper7944.html abstract	1-7, 9-12, 14-20, 22-29, 31-38, 40-48
Y	US 2005/0153985 A1 (Kehrli et al.) 14 July 2005 (14.07.2005) para [0011], [0052], abstract	8, 13, 21, 30, 39
Y	US 2010/0234452 A1 (Mian et al.) 16 September 2010 (16.09.2010)(para [0020], [0028], [0029], [0583])	8, 21, 39
Y		13, 30
☐ Further documents are listed in the continuation of Box C. ☐		
* Special categories of cited documents: "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family		
Date of the actual completion of the international search 29 October 2015 (29.10.2015)		Date of mailing of the international search report 08 DEC 2015
Name and mailing address of the ISA/US Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-8300		Authorized officer: Lee W. Young PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774