THESIS OF DOCTORAL (PhD) DISSERTATION

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A SURVEY ON THE USABILITY OF RADIO FREQUENCY BASED INDIVIDUAL IDENTIFICATION SYSTEM IN CASE OF DIFFERENT TYPES OF POULTRY

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1. INTRODUCTION

Currently, the traceability of food and its ingredients within the food supply chain comes as basic criteria. Knowing the origin of the products is not only a legal issue, but a need of consumers as well. In case of animals, tracing and tracking the product from farm to the consumers' table could only be possible with the use of individual tagging; while in the case of processed food products the solution is product marking. Animal identification ensures total traceability of the product from farm to consumers' table, which largely aids the decrease of food safety risks. Making tracking possible is essential in the poultry sector since the majority of food safety problems are generated by this sector. Currently, tracking of poultry in Hungary is handled by the Poultry Information System (PIS). The purpose of the system is to register the farms of breeding and farms of consumption, furthermore to track the geographical movement of the poultry consignments by documentation. As a drawback, PIS has an extremely high need of administration, which means the investment of more working hours. Following the examples of cow and sheep livestock, introducing radio frequency identification (RFID) based animal identification and tracking system for the poultry sectors could be advisable. By applying RFID system the electric traceability could be possible, which would make the procedures of the poultry sector more transparent. The aim of our study is to investigate the usability of radio frequency based individual identification method in the case of different types of poultry.

2. THE INVESTIGATIONS OF THE AUTHOR

2.1. The aim of the experiments

In case of cow, swine and sheep livestock developed RFID based individual identification methods are at our disposal. When it comes to poultry, however, the literature provides insufficient amount of information. How RFID identification affects production parameters, the physiological and stress state, or whether it causes any tissue irritation is hardly dealt with. That is why experiments, which are the topic of the dissertation, aimed at investigating the usability of RFID based identification in the case of different types of poultry. In order to determine it, tagging experiments were initiated, covering the following poultry types:

- broiler chickens
- turkeys
- geese
- ducks

The following issues were dealt with in case of every type:

- What effect RFID based identification has on the body weight and loss rate of tagged individuals?
- Does the applied individual tagging method affect the physiological (packed cell volume, aspartate aminotransferase, γglutamyltransferase, C-reactive protein concentration) and stress state (glucose and corticosterone content)?
- What readability percentage characterizes the applied RFID microchips, furthermore how is the durability of the used tagging method?

- Does tagging trigger any tissue irritation, alternation or inflammation on the site of implantation?

2.2. Materials and methods

Throughout the broiler chicken, turkeys, geese and ducks tagging experiment the feeding and watering were available *ad libitum*. In order to determine the effect of tagging on production parameters, the body weight of control and experimental animals was measured individually at the end of the certain feeding phases, furthermore the loss rate was recorded. Throughout the experiments we monitored the tagged animals for behavioral change, feather and tag pecking, and cannibalism-generated loss rate.

In order to determine physiological and stress state blood samples were taken in the case of every poultry types. Investigating RFID usability was extended to determining durability (loss percentage) and chip readability. The possible histological alternation on the site of implantation was monitored by histological examination.

2.2.1. Tagging experiments on broiler chicken

2.2.1.1. First group of experiments

420 COBB genotype broiler roosters were subjected to the broiler experiment initiated on the research farm of the Research Institute for Animal Breeding and Nutrition, Herceghalom. Two treatments were designed throughout the examination. The individuals of the experimental treatments were tagged with passive RFID microchip (MicroSensys GMBH, Erfurt, Germany), while the no individual tagging was used in the control

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group. The applied EM4135 type chips were working on 13.56 MHz, with 2 kbit data storage capacity. The communication speed was 26,4 kb/s, the readability range between 5-200 mm. The chips used during the experiments were regulated by the rules of the ISO 15693 standard. While tagging the individuals, the passive RFID chips were implanted into the patagium. During the experiments, the 1 day old birds were tagged in the doubled leather layer between the humerus and radius, (Fig 1).



Fig 1 The optimal poultry tagging site (A) radius, (B) ulna, (C) humerus, (X) implantation site.

The experiment was initiated with 8 replication in the case of the control group (240 chickens), and with 6 replication in the case of the experimental group (180 chicken). While in the hatchery, the individuals were vaccinated against Newcastle disease and bronchitis. The individuals were fed until their 42nd day. The animals were placed in colony brooders for 1 week, control and experimental individuals separated. From the 2nd week the tagged individuals were placed into 6, untagged animals into 8 deep litter pens with the bird density of 6 poults/m² (30 individuals/pens). Room temperature, humidity, ventilation, lighting program and number of dark

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hours were determined according to suggestions of the breeders' cooperative of hybrids. In case of both the experimental and the control groups a 3 phase feeding was used. The starting feed (weeks 0-1) was presented in morsel, the raising feed (2-4 weeks) and the finishing feed (5-6 weeks) in granulated form. In the broiler tagging experiment the 2008 suggestion of COBB was taken into consideration.

2.2.1.2. Second group of experiments

The aim of the second broiler tagging experiment (Research Institute for Animal Breeding and Nutrition, Herceghalom) was to determine the physiological background of possible behavioral change. 674 COBB broiler roosters were subjected to the experiment. The breeding and feeding circumstances were identical to the one applied during the first broiler tagging research. The control group consisted of 336, while the tagged group consisted of 338 individuals.

2.2.2. Tagging experiment on turkeys

The turkey tagging experiment was initiated on the plant of the Research Institute for Animal Breeding and Nutrition, Gödöllő. Hybrid XL genotype male turkeys were subjected for the purpose. The experiment was initiated with 9 replication in case of the control group (106 turkeys), and with 8 replication in case of the experimental group (94 turkeys). The individuals were fed from the 1st until their 19th day. After hatching the poults were vaccinated in the hatchery against Newcastle Disease and turkey rhinotracheitis, where debeaking with laser was also done. On the 21st day of the experiment re-vaccination against Newcastle disease; on the 28th day

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against turkey rhinotracheitis was made. The used tagging method was similar to the one discussed in the case of broilers. Tagged and untagged individuals were kept in colony brooders for 1 week. On the 7th day of the experiment birds from the tagged individuals were placed into 8, untagged into 9 deep litter pens with the bird density of 2 poults/m² (12 animals/pens). Room temperature, humidity, ventilation, lighting program and number of dark hours were determined according to suggestions of the breeders' co-operative of hybrids. A 6 phase feeding was used during the research. During the first phase (0-4th week) feed was presented in morsel form, during the later phases in 3 mm pellets. During the experiment on turkeys the 2010 suggestions of Gallicoop Ltd. were taken into consideration.

2.2.3. Tagging experiment on geese

Gourmaud liverhybrid geese were used for our geese tagging, initiated on the research plant of the Research Institute for Animal Breeding and Nutrition, Herceghalom. The experiment was initiated with 14 replications in case of both the control and experiment groups (196 geese in both groups). Individuals were fed from the 1st day until their 9th week. The used tagging method was similar to the one discussed in case of broilers. Birds were tagged on day 1 and both the tagged and control geese were placed into 14 deep litter pens, with 3 geese/m2 density (14 geese/pen, 7 ganders and 7 geese). The applied room temperature and lighting program was determined according to the suggestions of the breeders' co-operative. A 3 phase feeding was used, taking the 2004 suggestion of the Magyar Takarmánykódex. At the starting phase (0-4th week) feed was presented in

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coarse form, during the raising (4th-7th week) and finishing (8th-9th week) in granulated form.

2.2.4. Tagging experiment on ducks

Szarvasi K-94 hybrid ducks were used for our duck tagging. The experiment was initiated with 7 replications in case of both the control and experimental groups (126 individuals in the control and 132 in the experimental groups) on the research plant of the Research Institute for Animal Breeding and Nutrition, Herceghalom. Individuals were fed from the 1st day until their 7th week. The used tagging method was similar to the one discussed above. Birds were tagged on day 1 and both the tagged and control ducks were placed into 7-7 deep litter pens, with 3,5 duck/m² density (18 ducks/pen, 9 hens and 9 drakes). The applied room temperature and lighting program was similar to the geese experiments'. A 2 phase feeding was used, throughout the starting phase (0-4th weeks) feed was presented in coarse form, during the raising phase (3-7th week) in granulate. When planning the tagging experiment on ducks, the 2004 suggestions of the Magyar Takarmánykódex were taken into consideration.

2.2.5. Determining physiological and stress state of the examined poultry types

Blood samples were taken from the *vena cutanea ulnaris* of 10 tagged and 10 untagged individuals at the end of the experiment (42 days old broilers, 133 days old turkeys, 63 days old geese, and 49 days old ducks) for physiological and stress indication measurements. Literature concerning the effect of individual tagging is restricted to corticosterone content investigation. In order to gain more precise data of the physiological and

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stress tendencies, our examination was extended to further blood parameters. Blood samples (coagulation blocked with heparine) were analyzed for packed cell volume (PCV), aspartate aminotransferase (AST), γ -glutamyltransferase (γ -GT), C-reactive protein (CRP) and glucose concentration. The corticosterone (CORT) concentration was measured from blood plasma (coagulation blocked with EDTA). PCV was determined after separation, using a StatSpin centrifuge (10.000 RPM) (Iris Company, Norwood, USA). For the AST measurement, the reagent stock – catalogue number RANDOX AS 3804 (Randox Laboratories Ltd., Crumlin, UK) was used. For the γ -GT determination the RANDOX GT 3817 test kit was used. The C-reactive protein determination made with was immunturbidimetry method, with RANDOX CP 3826 kit was used. The glucose content was measured with glucose oxidase - peroxidase (GOD-POD) method with the RANDOX GL 3815 test kit. The AST, γ -GT, CPR, glucose determination was made with RANDOX RX Daytona (Randox Laboratories Ltd., Crumlin, UK) appliance. Determination of the blood plasma CORT concentration was used according to the procedures of the Csernus-style H-3 Corticosterone RIA (Radio Immuno Assay) kit (Csernus, 1981), using a Tri-carb 2800TR PerkinElmer Liquid Scintillation Analyzer (PerkinElmer Life and Analytical Science, Shelton, USA). In case of broiler chickens, the samples underwent heat treatment (56 °C 1h) before the CORT determination.

2.2.6. Examination of durability of tagging method, and readability percentage of microchips

Throughout the tagging experiments, the tag loss rate was determined at the end of each fattening phases when the body weight was measured. Furthermore, the patagium of the tagged individuals was monitored.

Readability was examined on the day of tagging and at the end of the experiment, with an RFID reader working on 13,56 MHz. The readability percentage of microchips was determined with the following formula.

 $R\% = \frac{\text{number of read RFID microchips}}{\text{number of used RFID microchips}} \times 100$

Calculating the readability rate (R%) (Caja és mtsai, 1999)

2.2.7. Methodology of the histological examination

Before the histological investigations 8-8 marked chickens, turkeys geese and ducks were slaughtered; part of the tissue around the implanted tag in the leather layer between the humerus and radius was extracted. The samples for histological research were fixed in 10% formaldehyde then samples were stained with heamotoxylin-eosine stain. The purpose of the investigations was to diagnose any possible histological alternation triggered by the marker.

2.2.8. Statistical evaluation

The statistical evaluation of the results was made with the use of SPSS 13.0 for Windows program (SPSS Inc., Chicago, USA). The

Kolmogorov-Smirnov test for normality of the data was used and, for normally distributed data we used the t-test to determine the significance between the measured parameters of the groups (Levene test, Independent Samples T test). In case of non-normal distribution, the significance was investigated with a non-parametric test (Mann-Whitney U-test). The chosen significance level in both cases was P<0,05.

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3. NEW SCIENTIFIC RESULTS

On the basis of the outcomes of our individual tagging experiments on broiler chickens, turkeys, geese and ducks the following scientific results can be stated.

- 1. Individual tagging with EM4135 type microchip equipped wing tags did not affect the body weight (measured at the end of fattening), the loss rate, the packed cell volume, and the aspartate aminotransferase, or γ -glutamyltransferase concentration of blood in the case of the examined poultry types (broiler chicken, turkeys, geese, and ducks).
- 2. The glucose and corticosterone concentration of blood plasma was not affected by RFID tagging in the case of broiler chickens and turkeys.
- 3. In case of tagged broilers, turkeys and ducks the average concentration of the inflammation indicating factor (C-reactive protein) showed no difference between the experimental and control groups. However, in case of tagged geese the concentration of C-reactive protein was significantly higher than of the average result of untagged individuals.
- 4. The outcomes of the histological research prove that patagial tagging did not cause local irritation, purulent inflammation an indicator of toxic effect cell necrosis, abscess generation or atypical cell sprouting in any of the poultry types.

5. The tag loss rate – an indicator of the durability of individual tagging method – exceeded the results of literature in the case of every poultry types. In order to increase durability of the tagging method – that is to say to be able to provide farm to consumers' table traceability – technological development of the construction of the tag would be needed.

4. LIST OF PUBLICATION MADE IN THE THEME OF THE DISSERTATION

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