A survey on frequency of infection to the virus of enzootic bovine leukosis in dairies around Tehran.

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Summary

Enzootic bovine viral leukosis not only can very much restricts cattle economics indifferent ways but also in the meanwhile investigation on the field of EBVL is be coming a model for oncology and studing some viral disease of mankind. So infection to the causative agent as well as the affection to the EBVL have been the subject of very many studies in most countries of the world. In IRAN for the first time by the use of AGIDT method we randomly tested 4797+130 serums of Holstein and native breed respectively. Results of this study indicates that a total of %9. 81 of the Holstein breed are serum converted while none of native breed are serum positive. In the infeted dairies the rate of infection ranges between %0.5-41.76. From 31 tested dairies with Holstein 26(83. 87) are infected. It is concluded that in IRAN enzootic

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bovine viral leukosis is an exotic disease and in the affected farms contact and therefore crowding are very important in spreading the infection. In the meantime there is a significant relation between age, milk yield and number of parturition on the one hand and the rate of infection to the causative virus on the other.

INTRODUCTION

Years before the recognition of the causative virus of EBVL, in some parts of the world especially in Europe contagious bovine leukosis was known and its control was planned and with some success applied (46,12,20).

In 1969 (1) for the first time, the causative virus of the disease was isolated and thereafter till now informations on the virus itself and viral leukosis has been gaining.

On the basis of present knowleg the virus of EBVL has worldwide distribution (37,5,35,42) and in the infected herd of Cattle is transmitted both vertically and horizontally (47,21,28,43,42,37,7,8,36,38,23,45,44,9,34,33,30,22,32 15).

A part from in breed susceptibility variation, the infection to EBL virus is much more prevalent in dairy than beef breed(36,7,8). In both breed, Cattle under the exposure of the virus either, in normal circumstances, genetically resists or infection stabilishes(7,18,15). In the latter case with some exception, after an incubation period of 4 years, the most important outcom
is the development of firm and white masses of malignant tumor in nearly all peripheral and internal organs and tissues with different frequency and severity (41, 7, 42, 41, 38, 29, 48, 42).

However, EBVL either by sudden death (acute form of leukosis or lymphosarcoma of adrenal glands, spleen and abomasum) or by the decrease of Production and reproduction performance and carcass condemnation (chronic type of leukosis) directly threatens economic life of cattle considerably.

In addition, economic effects of EBVL on the cattle (male or female) and sperm exportation would not be overlooked at all. Finally, it is worthwhile mentioning that the subject of leukosis to be a zoonosis is not yet dissolved (7, 15, 37, 44).

It is the above mentioned importance of leukosis that EBVL has been the subject of very many studies in most countries of the world and this survey is one of those but a very limited one.

Materials and Method

In this survey 4797 Serum samples of holstein friesian breed from 31 dairy herds which located around Tehran were tested. in the meanwhile 130 serum samples from an IRANIAN native breed called Sarabi which raised in the suburbs of karaj (a City of Tehran province) as the same manner as hostein fiesien breed were also tested. Map No. 1 shows the distribution of tested herds. with one
exception (herd No.6) all tested Cattle were above 2 years old. In the meantime in order to Search thoroughly, on the native farm at the second sampling, blood was taken from alarge numbers of one day old calves upto oldest Cows.

The test was done by the use of micro AGID method and the antigen was glicoprotein (gp) of EBL virus which was, in companywith the standard antiserum, supplied generously by Kono(Japan) and the procedure was as the same as Kono et al (22) and the suspected cases were re-tested.

The interval between sampling and testing the samples was, never, more than 5 days and during this time the samples were kept in the refrigerator at 4O c.

Results:

From 4797 samples 471 (9.81%) were serocon verted. 26 of 31 tested dairy farms were infected. This means that more than 83.87% of sampled herds were infected with EBL virus. In the infected herds the rate of infection varied between 0.5 and 41/76(Table 1). on the basis of this results, there is no relationship between the population of infected herds and the infection rate of them (table 2 and fig 1). the distribution of infected cattle among age groups and parturition numbers in infected herds are shown in figures 2 and 3. It appears that there is not any regular trend between the infection
rate and the either of the two above mentioned Parameters. Anyhow, Statistical informations on one of the infected farms showed that there is a significant (P 0.05) relationship between each of age, mean of daily milk yield, parturition numbers on the one hand and the infection rates on the other (table 3,4,5). In the meanwhile, on the same infected herd the duration between the time of Parturition and sampling date had no negative effect on the results of the test (table.6).

However none of the Cattle in native breed were seroconverted in two tests with an interval of at least 2 years between them. (Table,1)

Discussion

It is true that each of ELISA and RIA methods are more sensitive than AGIDT but on the basis of the present information the latter test has been well known as the standard one so for the identification of a herd infection rate is considered but not that of an individual Cow. Definetely doing AGIDT by the use of both p and gp Antigens gives more reliable results than each of the two. but because, 6 weeks after the establishment of infection, the majority of infected Cattle are seroconverted to gp antigen, it is now becoming an standard method to do AGIDT by making use of this antigen as we did(4,14,32, 39,10.3,24,26,16,22,19,40,15,).

The most strilking result of this survey was that
EBL virus has been introduced to IRAN by imported Cattle and the reasons for this claim are:

1- the infection is limeted to holstein friesian breed either imported or Iranian in breed of this Cattle which are raised in intensive or semi-intensive manner and never have been in contact with native breed.

2- the origin of the imported cattle are the countries where infection with EBL virus is quite common. 3-none of the Cattle in native breed which tested twice were seroconverted (table 1).

On the basis of this results it is clear that only in infected herd the intensivity and especially contact inbetween different age groups are the two very important factors in spreading the causative virus and increasing the rates of infection (table 2). This conclusion is in accordance with the findings of others in the one hand and on the other it indicates what means by the enzoocy of BVL (7,35,8,33,15,42).

The relationship between age and infection rates is well known (42,39, 7), and the two parameters, namely parturition numbers and daily milk yeild not only are closely related to age but also stresses resulted from high milk production and parturitions can make the cattle more sensitive to both infection and affection as it can be seen on table 3,4,5.

In our opinion, in infected herds application of hygienic regulations which difinitely increases mean of age groups can raise the infection rates. in addition unste-
rilized means for bleeding, injection, vaccination dehorning, tatooing and artificial insemination as well as the infected bull can transmitt EBL virus (15,36,25,26, 27,11,13,42,7,10) and increase the rates of infection. Because of these various tools of Transmission it should not be surprising to see high infection rates in either of well managed and populated farms or poorly managed and populated ones (table 2).

At the first look to figure 3 one might finde some what different from all mentioned above, but it is not so. This figure shows an unpleasant fact that the 2-3 yrs age group is dominant in nearly all herds and this is so because of not only tuberculosis and brucellosis are under eradication program but also the poor hygien and not to tackle properly with the diseases and affected animals. under such situation many cattle especially above 3 years old are culled.

Recommendations

In IRAN it is not economically wise to go through an eradication plan for leukosis bu the three following practicable points should be considered exactly:

1- Importation of Cattle (female and male) and sperum only from leukosis free herds should be permitted.

2- At least every 3 month serums collected for brucellosis eradication should go under the test of AGIDT and for the replacement purpose all seroconverted Cattle raised together and separate of the others.

3- The infected Herdsmen should not mixed attle of dif-
ferent age groups, should not make use of unsterilized means for bleeding, injection, vaccination, dehorning, artificial insemination and infected bull as well, should separate calves from their dams as soon as delivered and feed them with boiled, mixed clostrom and milk.
Map 1. Tehran Province and the Suburbs

Dotted areas were under survey
Table 1- Rate of infection to the causative agent of enzootic bovine viral leukosis

<table>
<thead>
<tr>
<th>Herd No</th>
<th>No. of Samples Per herd</th>
<th>No. Positive</th>
<th>Rate of Infection %</th>
<th>Herd No.of Samples Positive</th>
<th>No. of samples</th>
<th>Rate of infection %</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>63</td>
<td>10</td>
<td>15.78</td>
<td>17</td>
<td>9</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>49</td>
<td>1</td>
<td>16.33</td>
<td>18</td>
<td>102</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>40</td>
<td>3</td>
<td>7.5</td>
<td>19</td>
<td>198</td>
<td>1</td>
</tr>
<tr>
<td>4</td>
<td>31</td>
<td>2</td>
<td>6.45</td>
<td>20</td>
<td>278</td>
<td>1</td>
</tr>
<tr>
<td>5</td>
<td>167</td>
<td>32</td>
<td>19.16</td>
<td>21</td>
<td>44</td>
<td>4</td>
</tr>
<tr>
<td>*6(1)</td>
<td>241</td>
<td>0</td>
<td>0</td>
<td>22</td>
<td>29</td>
<td>0</td>
</tr>
<tr>
<td>*6(2)</td>
<td>73</td>
<td>0</td>
<td>0</td>
<td>23</td>
<td>210</td>
<td>58</td>
</tr>
<tr>
<td>7</td>
<td>139</td>
<td>6</td>
<td>4.32</td>
<td>24</td>
<td>153</td>
<td>35</td>
</tr>
<tr>
<td>8</td>
<td>314</td>
<td>21</td>
<td>6.69</td>
<td>25</td>
<td>91</td>
<td>38</td>
</tr>
<tr>
<td>*9(1)</td>
<td>225</td>
<td>11</td>
<td>4.89</td>
<td>26</td>
<td>30</td>
<td>11</td>
</tr>
<tr>
<td>*9(2)</td>
<td>89</td>
<td>7</td>
<td>7.86</td>
<td>27</td>
<td>50</td>
<td>1</td>
</tr>
<tr>
<td>10</td>
<td>252</td>
<td>20</td>
<td>7.94</td>
<td>28</td>
<td>50</td>
<td>6</td>
</tr>
<tr>
<td>11</td>
<td>188</td>
<td>34</td>
<td>18.08</td>
<td>29</td>
<td>50</td>
<td>1</td>
</tr>
<tr>
<td>*12(1)</td>
<td>470</td>
<td>0</td>
<td>0</td>
<td>30</td>
<td>16</td>
<td>0</td>
</tr>
<tr>
<td>*12(2)</td>
<td>325</td>
<td>17</td>
<td>5.23</td>
<td>31</td>
<td>118</td>
<td>30</td>
</tr>
<tr>
<td>13</td>
<td>43</td>
<td>2</td>
<td>4.65</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>14(1)</td>
<td>182</td>
<td>19</td>
<td>10.44</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>*14(2)</td>
<td>80</td>
<td>20</td>
<td>25</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>143</td>
<td>27</td>
<td>18.88</td>
<td>**32(1)</td>
<td>32</td>
<td>0</td>
</tr>
<tr>
<td>16</td>
<td>37</td>
<td>4</td>
<td>10.81</td>
<td>**32(2)</td>
<td>98</td>
<td>0</td>
</tr>
</tbody>
</table>

Total No. of Samples (Holstein) | Total No. of Positive | Mean rate of infection %
--------------------------------|------------------------|----------------------
4797                              | 471                    | 9.87                 |

* - Numbers in brackets represent repeadty of sampling in the same farms.
with an interval of at least 6 month

**- Native breed (sarabi) which, in this case raised as the same as Holstein Friesien.
Table 2- Certain farming parameters and infection rates to EBV in a herd infected with different sources of infection.

<table>
<thead>
<tr>
<th>No. of</th>
<th>Population of infected herd</th>
<th>Mean of Daily Milk yield/Cattle</th>
<th>Rate of infection %</th>
<th>Origin of cattle</th>
<th>Type of vaccination</th>
<th>Source of sperm</th>
<th>Situation of animal contact on each Herd</th>
</tr>
</thead>
<tbody>
<tr>
<td>20</td>
<td>1000</td>
<td>20.0</td>
<td>0.0</td>
<td>U.S.A</td>
<td>Natural</td>
<td>Hol. Inbr./1.p</td>
<td>Contact with same-age older ones</td>
</tr>
<tr>
<td>21</td>
<td>900</td>
<td>19.0</td>
<td>0.2</td>
<td>U.S.A</td>
<td>Natural</td>
<td>Hol. Inbr./1.p</td>
<td>Contact with same-age older ones</td>
</tr>
<tr>
<td>22</td>
<td>800</td>
<td>18.0</td>
<td>0.0</td>
<td>U.S.A</td>
<td>Artificial</td>
<td>Hol. Inbr./1.p</td>
<td>Contact with same-age older ones</td>
</tr>
<tr>
<td>23</td>
<td>600</td>
<td>20.0</td>
<td>0.0</td>
<td>U.S.A</td>
<td>Artificial</td>
<td>Hol. Inbr./1.p</td>
<td>Contact with same-age older ones</td>
</tr>
</tbody>
</table>

Notes:
- Holstein inbreeding/ Iranian product
- Common trough
- Calf up to first month of life maintained in a box
- Because of restriction of land, nearly all population being in contact directly with each other
- One month before parturition pregnant heifers and pregnant cattle live together.
### Table 3: Rate of infection in relation to Age

<table>
<thead>
<tr>
<th>Age (Year)</th>
<th>A</th>
<th>G</th>
<th>I</th>
<th>D</th>
<th>T</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 2</td>
<td>14</td>
<td>58</td>
<td>72</td>
<td>19.44</td>
<td>19.60</td>
<td>131</td>
</tr>
<tr>
<td>2 - 6</td>
<td>4</td>
<td>16</td>
<td>20</td>
<td>48.83</td>
<td>66.66</td>
<td></td>
</tr>
<tr>
<td>&gt; 6</td>
<td>11</td>
<td>13</td>
<td>24</td>
<td>66.66</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>39</td>
<td>92</td>
<td>131</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\[ x^2 = 1.731 \]

\[ x_3^2 \text{ and } 0.95 = 7.815 \]

(p < 0.05)
<table>
<thead>
<tr>
<th>Mean of Milk Yield (Kg/day/Cattle)</th>
<th>A</th>
<th>G</th>
<th>I</th>
<th>D</th>
<th>T</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 20</td>
<td>12</td>
<td>19</td>
<td></td>
<td></td>
<td></td>
<td>31</td>
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<tr>
<td>20-22.5</td>
<td>4</td>
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<td>22.5-25</td>
<td>3</td>
<td>18</td>
<td></td>
<td></td>
<td></td>
<td>21</td>
</tr>
<tr>
<td>&gt; 25</td>
<td>9</td>
<td>23</td>
<td></td>
<td></td>
<td></td>
<td>32</td>
</tr>
<tr>
<td>Total</td>
<td>28</td>
<td>90</td>
<td></td>
<td></td>
<td></td>
<td>118</td>
</tr>
</tbody>
</table>

$x^2 = 7.90$ and $0.95 = 7.815$ (P < 0.05)
Table 5-: rate of infection in relation to No. of Parturition

<table>
<thead>
<tr>
<th>No. of Parturition</th>
<th>A_+</th>
<th>G_+</th>
<th>I_-</th>
<th>D_-</th>
<th>T_-</th>
<th>Total</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>15</td>
<td>67</td>
<td></td>
<td></td>
<td></td>
<td>82</td>
<td>18.29</td>
</tr>
<tr>
<td>2</td>
<td>3</td>
<td>7</td>
<td></td>
<td></td>
<td></td>
<td>10</td>
<td>30.00</td>
</tr>
<tr>
<td>3</td>
<td>2</td>
<td>7</td>
<td></td>
<td></td>
<td></td>
<td>9</td>
<td>22.22</td>
</tr>
<tr>
<td>4</td>
<td>8</td>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td>11</td>
<td>72.72</td>
</tr>
<tr>
<td>&gt; 4</td>
<td>11</td>
<td>5</td>
<td></td>
<td></td>
<td></td>
<td>16</td>
<td>68.75</td>
</tr>
<tr>
<td></td>
<td>39</td>
<td>89</td>
<td></td>
<td></td>
<td></td>
<td>128</td>
<td></td>
</tr>
</tbody>
</table>

\[ x_4^2 = 26.26 \quad \quad \quad x_4^2 \text{ and } 0.95 = 9.488 \quad (P < 0.05) \]
Table 6.: rate of infection in relation to interval (Month) between parturition and sampling

<table>
<thead>
<tr>
<th>Interval between Parturition and sampling</th>
<th>A</th>
<th>G</th>
<th>I</th>
<th>D</th>
<th>T</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 2</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>2-4</td>
<td>12</td>
<td>19</td>
<td></td>
<td></td>
<td>31</td>
<td>31</td>
</tr>
<tr>
<td>4-6</td>
<td>15</td>
<td>21</td>
<td></td>
<td></td>
<td>36</td>
<td>36</td>
</tr>
<tr>
<td>6-8</td>
<td>6</td>
<td>21</td>
<td></td>
<td></td>
<td>27</td>
<td>27</td>
</tr>
<tr>
<td>&gt; 8</td>
<td>5</td>
<td>27</td>
<td></td>
<td></td>
<td>32</td>
<td>32</td>
</tr>
<tr>
<td>Total</td>
<td>38</td>
<td>89</td>
<td></td>
<td></td>
<td>127</td>
<td></td>
</tr>
</tbody>
</table>

\[ x^2_3 = 7.13 \quad x^2_3 \text{ and } 0.95 \quad \text{NS} \]
Fig. 1: Distribution of rate of infection (%) in relation to herds population.
Fig. 2: Distribution of rate of infection (%) in different age group of cattle.
Fig. 3: Distribution of rate of infection (%) in herds on the basis of No. of partition

Rate of infection (%)
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بداً علت که لدوزهری‌نتویک و پروس‌گونه نه تنها از ارزش اقتصادی گاو را به روش مختلف حاصل می‌کند و بلکه سبب انکه مطالعه پیرامون لدوزهری‌نتویک به‌عنوان مدالی برای مطالعات تومورشناسی و بیماری‌های ویروسی انسان بهبود آید. آنلودگی به ویروس عامل لدوزهری‌نتویک گاو و خود بیماری دریسپاری از کشورهای جهان به فراوانی و مورد مطالعه قرار گرفته است. در ایران برای اولین بار ماه‌گاه از روش AGIDT پروتاینی از ۱۳۵-۱۷۸۹۷ سرم به ترتیب از گاو نزدیک به‌سوی نزدیک به‌سیار به‌سیار می‌باشد. نتایج این مطالعه نشان می‌دهدکه ۰/۸۱٪ از گاو نزدیک به‌سیار به‌سیار می‌باشد. در گاودار و پزشکان آلودگی به مبتلایان مبتلایان تحت مطالعه (۱۸۱٪) ۶۱-۵/۰٪ منجر به‌سیار به‌سیار می‌باشد. این بررسی نتیجه‌گیری شده که در ایران لدوزهری‌نتویک و پروس گاو وارداتی به‌سیار به‌سیار به‌سیار به‌سیار می‌باشد. بنابراین تراکم در گسترش آلودگی نهایی اهمیت را دارد. نتایج این بررسی نشان داد که ارتباط بین سن میزان تولید این و تعداد زایمان‌های از باید طرف و نسبی آلودگی به ویروس عامل بیماری از طرف دیگر نظر آماری با اهمیت بود.