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**A Serologic Specificity Observed in Immunization of Cattle
with Wisent Red Cells**

**Serologiczna specyficzność obserwowana przy uodpornianiu bydła
czerwonymi krwinkami żubrów**

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[With 1 Fig. & 1 Table]

The research program of the Mammals Research Institute at Białowieża provides among others for general study of the effects of crossing of wisent with domestic cattle. The fairly ample experimental material (wisents, cattle, hybrids) collected so far also allowed to commence investigations on the antigenic blood properties of the tested individuals. This research aims primarily at the determination of relationship and differences between the antigenic blood properties in wisent and domestic cattle, and at the analysis of those properties on the basis of the antigen picture of hybrid blood. We are trying to answer the question whether lack of a crossing effect may be related to differences in antigenic blood properties of the individuals used in crossing, and whether similarity in antigenic blood properties may be considered as the cause of effects obtained in crossing. In the present preliminary communication we wish to signalize that in the course of investigations a phenomenon has been observed which may perhaps be considered as a serologically specific reaction to immunization of cattle with wisent red cell antigens.

Several individuals of various animals were immunized with the wisent "Pomian" red cells in January — February 1961, among them several heads of domestic cattle from the Popielno herd, including the cow "Odra" No. 98 (now used for crossbreeding with wisent at Białowieża).

In the sera of immunized animals were found antibodies of various kinds against red cell antigens of the wisent-donor and also other wisents

as well as against red cell antigens of domestic cattle. This occurred also in the serum of the cow "Odra" No. 98. Antibody titre, determined by hemolytic tests, varied for different animal species within the range of 1:32 to 1:256. After accomplishing suitable absorptions we succeeded in isolating from the immune serum of cow "Odra" No. 98 antibodies reacting with blood antigenic factor X_1 (common for cattle and wisent) and with antigens specific for wisent.

About 2 years after the above mentioned tests, investigations were commenced (Sept. 1962) on the antigenic blood properties of particular individuals of the Białowieża experimental group. Red cells and sera of 6 cows and 3 hybrids of F_1 (Table 1) were used in various kinds of initial

Table 1.
Material used for experiments

No.	Species and name of animals	No. of animals
1.	Wisents: 1. Pomian (by Ponury, No. 902, from Polatucha, No. 832) b.... XI. 1960. 2. Pokorny (by Ponury, No. 902, from Pogoń, No. 827) b. 6.VII.1957.	2
2.	Cows of polish black and white lowland breed (Nos. 78, 98, 106, 107, 114, 117). Hybrids of wisent and domestic cattle (F_1)*	6
3.	1. Facet (by Pokorny, No. 1077, from cow No. 98) b.28.IV.1962. 2. Farad (by Pokorny, No. 1077, from cow No. 107) b.29.IV.1962. 3. Fanny (by Pokorny, No. 1077, from cow No. 114) b.30.IX.1962.	3

* For details dealing with crossings of wisent with cattle see K r a s i ń s k a (1963).

serological tests. During those experiments the presence of antibodies (hemolysins) was stated in the blood serum of the cow "Odra" No. 98. This serum reacted with red cells of a few individuals of the tested cattle with the red cells of every tested wisents and of hybrids, among them also of offspring of the cow "Odra" No. 98 — "Facet". In hemolytic reaction the titre of the antibodies contained in these sera varied in those tests between 1:32 and 1:64, according to the different kinds of red cells.

During a period of over 2 years (February 1961 — April 1963) the cow No. 98 did not receive any injection of foreign substances, except coffein immediately after the birth of the first hybrid-calf — "Facet".

The results of hemolytic tests of wisent, cattle and hybrids red cells with particular serum samples from the cow "Odra" No. 98, taken at six different dates of a 7-months' period, are presented in the enclosed diagram (Fig. 1).

The results of hemolytic tests show that every tested serum sample from the cow No. 98 contains antibodies against the cell antigens of cattle, wisent and hybrids. Only minor differences occur between serum samples taken at different dates of the 7 months' period in regard to reaction intensity (titre) of the antibodies. Also the intensity of the hemolytic reaction to particular red cell kinds showed no major variance. The

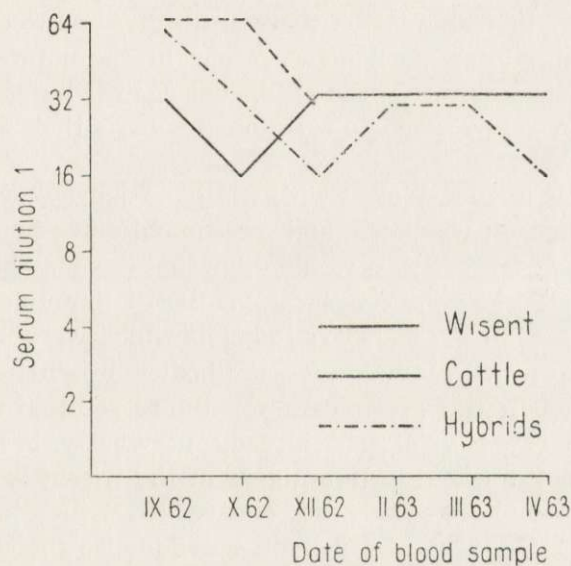


Figure 1. Hemolytic reaction of the blood serum of cow. No. 98 with the red cells of wisents, cattle and hybrids.

particular sera reacted in every case with the blood cells of the hybrid "Facet", offspring of the cow which serum was tested.

In order to determine the quality of the antibodies contained in the tested serum, some serum samples were subjected to absorption with various kinds of red cells from cattle or wisent. In the course of these experiments we succeeded in isolating antibodies reacting with antigenic factor X_1 , common for cattle and wisent (G a s p a r s k i, 1961). Any of the cattle red cells with the factor X_1 when used in absorption removed the corresponding antibodies in every case, but some fractions of antibodies reacting with the red cells of wisent and hybrids remained in the serum. The red cells of the tested wisent and hybrids removed in every case the

antibodies against the cell antigens of their own blood, and also those against the antigens of cattle blood.

The phenomenon observed and the related results of serological tests suggest certain reflections and allow to propose some working hypotheses.

As a result of immunization of the cow No. 98 with wisent red cells a "continuous production" of antibodies against certain selected antigens, (in the given case factor X₁), took place and against some antigens specific for wisent.

More data ought to be obtained regarding production intensity of the above mentioned antibodies in particular seasons of the annual cycle. It will then become possible to form a picture of eventual variations in reaction intensity (titre amount) of these antibodies and to compare them with those observed for particular seasons in the naturally occurring antibodies of cattle serum (e.g. the antibodies against the so-called J cell antigen — Stone, 1956), and to state on this basis their similarities or differences.

The crossings of the cow No. 98 with wisent "Pokorny" were effective. A hybrid calf "Facet", was born and presumably another embryo was aborted (Krasinska, 1963). It would seem that this may have caused — probably through the embryo — a "natural" immunization against the antigens present in the embryo's red cells which were inherited from the wisent "Pokorny". Production of antibodies by "natural" immunization could occur in the given case for the reason that the mother of the hybrid (the cow No. 98) had already previously been effectively immunized against the red cell antigens of the wisent "Pomian". "Pomian" and "Pokorny" are very near relatives since they have the same father ("Ponury", No. 902) who in both cases is also their grand-father from the maternal side. It may therefore be surmised that possible "natural" immunization had been produced by almost identical blood.

Quite apart from the question which of the two hypotheses discussed above may find confirmation in further research, the observed phenomenon suggests another problem — namely whether all the normally occurring antibodies against particular red cell antigens which have been detected so far can be really considered as "normal physiologic" (Neimann-Sorensen, 1958; Stone & Miller, 1953). If we assume for instance that we do not know the story of the immunization of cow No. 98 nor the properties of her serum hemolyzing the red cells of her own progeny then the observed antibodies in incidental tests of her serum, shall be considered as "normal physiologic" antibodies.

Investigations aiming at the elucidation of causes of the phenomenon of serologic specificity are being continued. To this end the amount of

experimental material shall be increased and a series of special tests performed, based on certain models from the research work by Kiddy et al. (1958) and by Menge et al. (1962), dealing with immunologic investigation of the problem of fertility and sterility in different animal species in its general aspect.

SUMMARY

In January—February 1961 cow "Odra" No. 98 was immunized with red cell of wisent "Pomian". From the immunized cow serum antibodies against antigenic factor X₁, and against antigens specific for wisent were isolated. In April 1962 cow No. 98 was mated with wisent "Pokorny" (No. 1077), having a similar blood type as "Pomian". The hybrid "Facet" was born and her subsequent gestation was interrupted probably by abortion.

Tests of her blood made during 7 month since September 1962 showed the presence of similar antibodies as those observed after immunization with the red cells of "Pomian". The serum of cow No. 98 also reacted with the red cells of her son "Facet".

It would seem that in cow No. 98 occurred a continuous production of antibodies or "natural" immunization through the embryo and against its red cell antigens, inherited from wisent "Pokorny".

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STRESZCZENIE

W okresie styczeń—luty 1961 r. uodporniono krowę nr 98 „Odra” krwinkami żubra „Pomiana”. Z odpornościowej surowicy tej krowy wyodrębniono przeciwciała przeciw czynnikom antygenowym X_1 (wspólnym dla żubrów i bydła) oraz antygenom krwinkowym specyficznym dla żubrów. W kwietniu 1962 roku w wyniku skrzyżowania z żubrem „Pokornym”, 1077 (który ma podobny skład antygenowy krwi jak żubr „Pomian”) krowa nr 98 urodziła hybryda „Faceta”, a następną ciążę prawdopodobnie poroniła. Badania jej krwi prowadzone ponownie od września 1962 r. przez 7 miesięcy wykazały obecność podobnych przeciwciał, jak po uodpornieniu krwinkami „Pomiana”. Surowica krowy nr 98 reaguje również z krwinkami syna — „Faceta”.

Wydaje się, że u krowy nr 98 prawdopodobnie wystąpiło trwałe wytwarzanie przeciwciał, lub „naturalna” immunizacja poprzez płód przeciw antygenom krwinek płodu, który oddziedziczył je od żubra „Pokornego”.

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