

1 **Title: Maintenance of insulin secretion capacity and reduction of GAD**  
2 **autoantibody titer in a patient with LADA: A case report<sup>1</sup>**

3  
4 **Running title: Treatment of LADA with metformin**

5  
6 **Keywords: glutamic acid decarboxylase autoantibody, insulin secretion capacity, latent**  
7 **autoimmune diabetes of adults, metformin**

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9 **Highlights:**

- 10  
11 • LADA patients with high GAD antibody titers typically require insulin treatment  
12 • A patient with high GAD antibody titer and intact insulin secretion refused insulin  
13 • Metformin reduced antibody titer and maintained 5-year insulin secretion capacity

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**<sup>1</sup>Abbreviations**

CPR, connecting peptide immunoreactivity; GAD, glutamic acid decarboxylase; GADAb, anti-GAD antibodies; IA-2Ab, insulinoma-associated antigen-2 antibody; IAA, insulin autoantibody; LADA, latent autoimmune diabetes of adults; SU, sulfonylurea; TgAb, thyroglobulin antibody; TPOAb, thyroxine peroxidase antibody; TPOAb, thyroxine peroxidase antibody; TZD, thiazolidinedione

**Commented [A1]:** Dear author, I have carefully read your manuscript on the use of metformin for treating a LADA patient with high GADAb but an intact insulin secretion capacity. Overall, it was clear and logically structured. I have made changes to improve language, structure, and submission readiness. I have also provided suggestions to strengthen the presentation of your research and improve logical flow. I hope you will find this useful.

16  
 17 ~~The~~ ~~We~~ report the case ~~is of a 52-52-years-year-old man-He who~~ was first diagnosed with diabetes  
 18 mellitus ~~with-based on~~ HbA1c 7.9% (65.2 mmol/mol) ~~by a medical examination for the first time~~  
 19 ~~in 2004~~. His ~~insulin secretory-secretion~~ capacity ~~of insulin~~ was ~~maintained-intact~~ (fasting serum  
 20 ~~connecting-peptide immunoreactivityC-peptide (CPR)~~ 2.5 ng/ml), but ~~the islet-autoantibody tests~~  
 21 ~~revealed against showed high~~ glutamic acid decarboxylase (GAD) ~~-autoantibody titers was high~~  
 22 (23.9 U/ml). ~~His HLA haplotype was testing revealed a~~ combination of susceptible ~~haplotype~~  
 23 (DRB1\*0405/DQB1\*0401) and protective ~~haplotype~~ (DRB1\*0803/ DQB1\*0601) ~~haplotypes~~. We  
 24 ~~started-initiated~~ metformin therapy ~~because we obtained no agreement foras he refused~~ insulin  
 25 therapy. ~~After metformin administration~~ ~~Thereafter, we observed reducinghis~~ GAD ~~-autoantibody~~  
 26 ~~from high titer to low titer titer decreased to~~ (1.7 U/ml) ~~- and hHis insulin secretory-secretion~~  
 27 capacity ~~of insulin~~ was maintained for ~~five 5~~ years. When ~~insulin secretory-secretion~~ capacity ~~of~~  
 28 ~~insulin is maintained-intact in patients with~~ latent autoimmune diabetes of adults (LADA) ~~ease~~  
 29 ~~with who have high GAD-autoantibody titers, it is possibly suggested that treatment by metformin~~  
 30 ~~administration istreatment may be~~ effective.

### 34 ~~{Introduction}~~

35 ~~Latent autoimmune diabetes of adults (LADA) is the-a condition in which ease that islet-related~~  
 36 ~~autoantibody-autoantibodies becomes prolonged positiveare present for a prolonged period,~~  
 37 ~~similar to type 1 diabetes, and-but in which the insulin secretion capacity is intact secretory~~  
 38 ~~capacity of insulin is maintained like type 2 diabetes mellitus at the time of onset, but iThis~~  
 39 ~~capacity reduces fordecreases over a period of several months to years, and-finally progressing to~~

**Commented [A2]:** It is unusual to start an abstract of a case report with a case presentation without presenting some rationale for the report. I suggest that you include a sentence here to lay the background for this report before presenting the case, in the line of the information you present in the Introduction: "The effect of oral anti-diabetes agents for treatment of patients with latent autoimmune diabetes of adults who have high glutamic acid decarboxylase autoantibody titers but intact insulin secretion capacity has not been established to date."

**Commented [A3]:** In the abstract, this information is not relevant, as this is the only date given.

**Commented [A4]:** Please check if the target journal allows use of this abbreviation without definition.

**Commented [A5]:** I notice that, in the text, you preferentially use the term "BG." I suggest that you use consistent terminology throughout the paper.

**Commented [A6]:** Define non-standard abbreviations at the first instance of the term in each part of the manuscript that should be able to stand alone, such as the abstract, main text, and each figure legend and table, unless otherwise directed by the journal.

**Commented [A7]:** Do you perhaps rather mean type 1 diabetes mellitus, which is associated with islet-related autoantibodies, whereas type 2 is not typically associated with such antibodies?

**Commented [A8]:** Descriptive phrases should be kept close to the terms they are describing. Here, I assume that you are referring to the similarity between LADA and diabetes in terms of the presence of islet-related autoantibodies, and have moved it accordingly.

**Commented [A9]:** When sentences are overlong, they become difficult to read. Rather split long sentences (more than about 50 words) where the topic shifts to improve readability.

a state of insulin-insulin-dependence.

The measurement of the anti-GAD antibody antibodies (GADAb) is most useful in for LADA diagnosis of disease, and it is known to progress beta-cells destruction and lead to insulin dependence easily when GADAb shows high titers (more than exceeding 10 U/ml) are indicative of beta-cell destruction, which is readily followed by insulin-dependence.<sup>1)</sup> We-In the Tokyo study, we have been reported that insulin administration inhibited progression to insulin-dependence better than did sulfonylurea (SU) therapy in LADA cases with high GADAb titers (more than exceeding 10 U/ml) and but with intact maintained-insulin secretion capacity, insulin administration is inhibited progress to insulin dependence compared with sulphonylurea (SU) therapy in Tokyo study.<sup>2)</sup> However, the effect of using other oral anti-diabetes agents for treatment therapy of other oral anti-diabetes agents for of LADA with high GADAb titers such patients in the stage of insulin independence has not been is not established now to date.

We present our experienced-experience of a LADA case with high GADAb titers that who refused insulin therapy and was treated effectively by with biguanide (BG) (Metformin/metformin) (biguanide) only, during for five-5 years because it is no agreement with insulin therapy. With this treatment, this case is patient's maintained insulin secretary secretion capacity was maintained of insulin for five-5 years; and reduced-his GADAb titers decreased markedly to low titers by BG administration. For the first time, wThis case highlights the reported this case because it showed value of BG-metformin therapy in-for LADA case-patients with maintained-intact insulin secretion capacity.

#### {Case report}

A 52-52-years-year-old man was diagnosed with diabetes mellitus, with-based on HbA1c 7.9% (65.2 mmol/mol), by during a company-based medical checkup/medical examination in his

**Commented [A10]:** This was somewhat awkwardly phrased. Please do check if my edit conveys your intended meaning accurately.

**Commented [A11]:** Insert a space between the number symbol and the unit of measurement.

**Commented [A12]:** When a noun (cell) is used as an adjective to describe another noun (destruction), use the adjectival noun in the singular (cell) rather than plural (cells) form.

**Commented [A13]:** Please do check that the citations are given in the style required by the target journal.

**Commented [A14]:** In US English, the "ph" diphtong is not used for the "f" sound.

**Commented [A15]:** As this is the generic drug name, rather than the drug brand name (which would be a proper noun), it should not be written with an upper-case first letter.

64 ~~company\_on\_in~~ September, 2004. He visited ~~saitama-Saitama soeial-Social insurance-Insurance~~  
 65 ~~hospital-Hospital~~ ~~to\_for\_treat~~ diabetes mellitus ~~treatment~~. ~~He has not been pointed out of~~  
 66 ~~h~~Hyperglycemia and glucose ~~intolerance~~ ~~abnormalitywere not identified~~. He ~~was suffered from~~  
 67 ~~had a history of~~ duodenal ulcer and acute hepatitis A ~~in\_at the age of~~ 21 years-old. ~~Questioning~~  
 68 ~~revealed that hisHis~~ parents ~~have\_had~~ type 2 diabetes mellitus. He ~~is\_nohad no~~ ~~history-of~~ smoking  
 69 ~~history, and-but reported~~ drinking beer (500 ml ~~every daydaily~~).

**Commented [A16]:** As this is a proper noun (the name of an institution), each word should start with an upper-case letter.

**Commented [A17]:** This sentence was somewhat unclear. Please check if my edit conveys your intended meaning accurately, and get back to me with an explanation if necessary, so that I can help you to phrase your meaning accurately but clearly.

70 A ~~physical examination revealed as follows~~. His height was 164.4 cm, ~~and his,~~ weight  
 71 was 71.0 kg (~~Max-maximum:~~ 74.0 kg at ~~age~~ 50 years-old). ~~His,~~ body mass index (~~BMI~~) was 26.3  
 72 kg/m<sup>2</sup>. ~~His,~~ blood pressure was 126/64 mmHg, and ~~his~~ pulse was 72 ~~beats/min~~. ~~He-Physical~~  
 73 ~~examination yielded showed no abnormal-remarkable~~ findings in physical examination.

**Commented [A18]:** I suggest that you move this sentence to the start of this paragraph to improve flow.

74 A ~~l~~laboratory ~~examination-tests~~ revealed as follows. His fasting blood sugar (~~FPG~~)  
 75 ~~was\_of~~ 8.9 mmol/l, ~~and and~~ HbA1c ~~was\_of~~ 7.8% (64.2 mmol/mol) at ~~first the initial~~ examination.  
 76 His ~~insulin secretory-secretion~~ capacity ~~of insulin~~ was ~~maintained-intact~~ (~~Fasting-fasting~~ blood  
 77 ~~connecting peptide immunoreactivity [CPR]CPR~~ 2.5 ng/ml). ~~His fat metabolism was high levels~~  
 78 ~~of His~~ total cholesterol (~~TC~~) (261 mg/dl) and triglyceride (TG) (185 mg/dl) ~~levels were high~~. ~~His~~  
 79 ~~islet-related~~ autoantibody ~~tests~~ showed high GADAb titers (23.9 U/ml), but insulinoma-  
 80 associated antigen-2 antibody (IA-2Ab), insulin autoantibody (IAA), and zinc transporter 8  
 81 antibody (~~ZnT8Ab~~) ~~were negative both tests were negative~~. ~~His+I~~ thyroid-related autoantibody ~~tests~~  
 82 ~~showed\_were~~ positive ~~for~~ thyroxine peroxidase antibody (TPOAb) (>50.0 U/ml), ~~and-but~~ negative  
 83 ~~for~~ thyroglobulin antibody (TgAb). ~~His~~-HLA ~~was\_testing revealed a~~ combination of susceptible  
 84 ~~haplotype~~ (DRB1\*0405/DQB1\*0401) and protective ~~haplotype~~ (DRB1\*0803/DQB1\*0601)  
 85 ~~haplotypes~~. ~~His-He had no diabetic-diabetes-associated~~ complications, such as retinopathy,  
 86 nephropathy, and neuropathy ~~were not shown~~.

**Commented [A19]:** Do not abbreviate terms that are not used repeatedly in the text.

**Commented [A20]:** Do not abbreviate terms that are not used repeatedly in the text.

**Commented [A21]:** Insert a serial comma before the conjunction (and) that precedes the last item in a list (neuropathy).

87 His clinical course ~~was~~ shown in Figure 1. ~~At the initial examination, we~~ ~~We observed~~  
 88 ~~with~~ ~~did not prescribe~~ medication, ~~but~~ ~~and~~ ~~guide him in~~ ~~advised~~ -calorie restriction (1600 kcal  
 89 ~~per day)~~ ~~at first examination,~~ ~~but it showed elevation of~~ ~~However,~~ ~~his~~ HbA1c value ~~was~~ ~~elevated~~.  
 90 We recommend ~~him for~~ insulin therapy, ~~because it was the case with~~ ~~given that his~~ ~~high~~ GADAb  
 91 titer ~~was~~ ~~high~~ s (23.9 U/ml), ~~but it is obtained no agreement~~ ~~but he refused~~. ~~So,~~ ~~Consequently,~~ we  
 92 ~~started~~ ~~prescribed~~ BG (metformin (500 mg/day) ~~therapy from in~~ May, 2005. His glycemc control  
 93 ~~improved the first half of to~~ ~~7% in~~ HbA1c ~~7%~~; and his ~~insulin secretory~~ ~~secretion~~ capacity ~~of insulin~~  
 94 was ~~maintaining~~ ~~maintained~~ (fasting blood CPR: about 2 ng/ml ~~in~~ ~~fasting~~ ~~CPR~~) during ~~the~~ ~~five~~ ~~5~~  
 95 years ~~after~~ ~~following~~ ~~BG~~ ~~metformin~~ ~~administration~~ ~~initiation~~. Furthermore, ~~his~~ GADAb titers  
 96 ~~deteriorated remarkably~~ ~~reduced markedly~~ and ~~were~~ maintained ~~in~~ ~~at~~ low titers ~~afterward~~ ~~levels~~.  
 97 ~~His~~ ~~No~~ ~~diabetic~~ ~~diabetes-related~~ complications ~~such as~~ ~~retinopathy,~~ ~~nephropathy~~ ~~and~~ ~~neuropathy~~  
 98 ~~were not shown during~~ ~~developed over these~~ ~~five~~ ~~5~~ years.

**Commented [A22]:** Please indicate the time-period for such restriction.

**Commented [A23]:** Please indicated whether you are referring to the initial examination, or whether you mean that his HbA1c levels remained high over a particular observation period.

**Commented [A24]:** Scientific writing tends to be formal. Rather use the more formal "consequently" or "thus" than the informal "so".

**Commented [A25]:** It is not clear what you mean here. Do you mean that his HbA1c levels improved to 7% during the first half-year? Please clarify this or get back to me with an explanation so that I can help you to phrase this more clearly.

**Commented [A26]:** I suggest that you also mention whether the TPOAb title was in any way affected by this treatment.

**Commented [A27]:** As you have mentioned these previously, you could delete the details here.

**Commented [A28]:** I suggest that you clarify if this is the length of time that the patient was followed-up: was this status maintained at the time of writing?

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#### 100 **{Discussion}**

101 We ~~here~~ reported ~~the~~ ~~a~~ LADA case with high GADAb titers at onset ~~that~~ ~~who~~ ~~was~~ treated with ~~BG~~  
 102 (metfor-min) ~~therapy,~~ ~~not with~~ ~~rather than with~~ insulin ~~therapy~~ for ~~five~~ ~~5~~ years. ~~In this case,~~ ~~t~~ ~~The~~  
 103 ~~patient's~~ glycemc control was stable ~~with the first half of,~~ ~~with~~ HbA1c ~~7%~~ ~~in~~ HbA1c, and ~~the~~ ~~,~~  
 104 ~~and his insulin secretion~~ ~~secretory~~ capacity ~~of insulin~~ was maintain-ed ~~during~~ ~~five~~ ~~over~~ ~~the~~ ~~5-~~  
 105 ~~years~~ ~~year~~ ~~period~~. Furthermore, ~~the~~ ~~his~~ GADAb titers ~~decreased~~ ~~to~~ ~~low~~ ~~titers~~ ~~at~~ ~~an~~ ~~early~~  
 106 ~~stage~~ ~~markedly~~ ~~soon~~ after ~~starting~~ ~~BG~~ ~~metformin~~ administration.

107 High GADAb titers (~~more than~~ ~~exceeding~~ 10 U/ml)<sup>-2</sup>; and ~~positive~~ ~~positivity~~ for ~~in~~  
 108 several islet-associated ~~autoantibody~~ (GADAb<sup>-1</sup>, IA-2Ab<sup>-1</sup>, IAA)<sup>-3</sup>; ~~positive~~ ~~in~~ ~~and~~ thyroid-related  
 109 ~~autoantibody~~ (TgAb<sup>-1</sup>, TPOAb)<sup>-4</sup> ~~autoantibodies are suggested~~ ~~indicate~~ ~~as~~ ~~high-~~ ~~risk~~ ~~group~~  
 110 ~~progressing of disease~~ ~~progression,~~ ~~with~~ ~~decreased~~ ~~of~~ ~~the~~ ~~insulin~~ ~~secretory~~ ~~secretion~~ capacity ~~of~~

insulin-in LADA cases. ~~On the other hand, it is thought that decrease of the~~ However, the insulin secretory secretion capacity is ~~thought to be~~ delayed ~~when we have~~ in the context of a protective HLA ~~protective~~-haplotype. ~~This Our~~ case had a high GADAb titers and ~~was thyroid-related antibody (TPOAb)-~~ positive, but ~~it~~ had a ~~protective~~ HLA ~~protective~~-haplotype. ~~There is no report for evaluating~~ The degree of contribution ~~in of~~ each factor ~~toward~~ decreasing ~~ing of the~~ -insulin secretory secretion capacity ~~remains unclear~~. ~~But~~ We ~~considered~~ ~~surmised~~ that the presence of the ~~protective~~ HLA ~~protective~~-haplotype ~~is possibly~~ may have made a greater contribution ~~to progress insulin deficiency than the~~ than high GADAb titers and presence of ~~thyroid-related autoantibody~~ TPOAb, because it is ~~maintained~~ maintaining the patient's insulin secretory secretion capacity for a long time ~~in this case~~.

~~Now~~ Currently, insulin therapy is ~~firstly recommended for~~ the first-line treatment ~~of for~~ glycemic control in LADA cases, ~~because insulin administration is inhibited progression to as it inhibited progression to insulin~~ insulin-dependence, as compared ~~with to~~ SU therapy, in the Tokyo study.<sup>2)</sup> ~~It remains unclear about~~ The ~~underlying~~ mechanism ~~to inhibit progression to insulin deficiency in detail~~ remains unclear, but we ~~supposed~~ consider that it ~~inhibits~~ ~~destruction of~~ beta-cell ~~destruction~~ function by inducing ~~of~~ autoimmune tolerance and removing ~~an~~ excessive burden ~~for from the~~ beta-cells ~~by using insulin~~. In LADA cases, ~~it is reported that~~ SU therapy ~~should not use is not recommended~~.<sup>5)</sup> ~~and w~~ We thought that glinide therapy ~~is should~~ also ~~not recommended because it causes~~ be avoided, as it may ~~possibly progression to further~~ decrease the insulin secretory secretion capacity by increasing ~~burden for the insulin secretion burden on~~ beta-cells ~~to promote insulin secretion like, similar to~~ SU therapy. ~~On the other hand,~~ Thiazolidinedione (TZD) therapy, ~~which that improved~~ improves insulin resistance and decreases excessive insulin secretion, ~~is possibly~~ may be effective in LADA cases, ~~but the~~ although its influence ~~of on~~ immune

**Commented [A29]:** Scientific writing tends to be formal, whereas starting a sentence with "but" is informal. Rather replace "but" with the more formal "However" at the start of a sentence.

134 function (~~possibility to change affecting the~~ Th1/Th2 balance) ~~is supposed about it~~ may need to  
135 ~~be considered.~~ ~~There are various reports about the effect of using~~ Although the use of TZD has  
136 ~~been reported,~~ and it remains unclear ~~theits~~ effectiveness ~~of TZD therapy for in~~ LADA cases  
137 ~~remains unclear.~~<sup>6)</sup> We used ~~BG (metformin)~~ therapy ~~in this patient who refused insulin therapy,~~  
138 because it ~~improved~~ improves insulin resistance and ~~it suggested possibility to~~ may inhibit T-cell  
139 ~~cell-~~mediated immune responses.<sup>7)</sup> ~~in this case with no agreement for insulin therapy. There is not~~  
140 ~~reported that~~ No previous LADA cases ~~using in whom BG metformin~~ therapy was used for a long  
141 time ~~have been reported, previously. Sand it is thought that we will require the accumulation of~~  
142 similar cases should be accumulated in future.

**Commented [A30]:** This was not quite clear. Please check if my edit conveys your intended meaning accurately, or get back to me with an explanation so that I can help you to phrase your meaning more clearly.

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